Phytoremediation of Heavy Metal-Contaminated Sites: Eco-environmental Concerns, Field Studies, Sustainability Issues and Future Prospects

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Abstract

Environmental contamination due to heavy metals (HMs) is of serious ecotoxicological concern worldwide because of their increasing use at industries. Due to non-biodegradable and persistent nature, HMs cause serious soil/water pollution and severe health hazards in living beings upon exposure. HMs can be genotoxic, carcinogenic, mutagenic, and teratogenic in nature even at low concentration. They may also act as endocrine disruptors and induce developmental as well as neurological disorders and thus, their removal from our natural environment is crucial for the rehabilitation of contaminated sites. To cope with HM pollution, phytoremediation has emerged as a low-cost and eco-sustainable solution to conventional physico-chemical cleanup methods that require high capital investment and labor alter soil properties and disturb soil microflora. Phytoremediation is a green technology wherein plants and associated microbes are used to remediate HM-contaminated sites to safeguard the environment and protect public health. Hence, in view of the above, the present paper aims to examine the feasibility of phytoremediation as a sustainable remediation technology for the management of metals-contaminated sites. Therefore, this paper provides an in-depth review on both the conventional and novel phytoremediation approaches, evaluate their efficacy to remove toxic metals from our natural environment, explore current scientific progresses, field experiences and sustainability issues and revise world over trends in phytoremediation research for its wider recognition and public acceptance as a sustainable remediation technology for the management of contaminated sites in 21st century.

Keywords: Heavy metals, Trophic transfer, Environmental pollution, Toxicity, Hyperaccumulators; Phytoremediation; Phytoextraction, Phytovolatilization, Phytostabilization, Rhizodegradation, Rhizofiltration, Molecular mechanism; Plant growth promoting rhizobacteria; Endophytes; Arbuscular mycorrhizal fungi; Engineered plants; Halophytes, Aromatic plants, Energy crops; Phytotechnologies; Phytomining; Agromining; Constructed wetlands; Contaminated sites; Field studies

1 Introduction

Environmental pollution is of serious ecological concern worldwide with a continually rising public outcry to ensure the safest and healthiest environment. A variety of organic and inorganic pollutants have been reported to cause environmental pollution and severe health hazards in living beings (Maszenan et al. 2011; Saxena and Bharagava 2017). Among them, heavy metals (HMs) are highly notorious pollutants due to their high abundance and nonbiodegradable and persistent nature in the environment. Hence, they cause soil/water pollution and toxic, genotoxic, teratogenic and mutagenic effects in living beings (Dixit et al. 2015; Sarwar et al. 2017). They also cause endocrine disruption and neurological disorders even at low concentration (Yadav 2010; Maszenan et al. 2011; Dixit et al. 2015; Sarwar et al. 2017). Any naturally occurring metal/metalloid having an atomic number greater than 20 and elemental density greater than 5 g cm^{-3} is termed as HM. They includes copper (Cu), cadmium (Cd), chromium (Cr), cobalt (Co), zinc (Zn), iron (Fe), nickel (Ni), mercury (Hg), lead (Pb), arsenic (As), silver (Ag) and platinum group elements (Ali et al. 2013; Ali and Khan 2018a). Among them, Cd, As, Hg, and Pb don't have any biological function in the body and thus, are non-essential elements. They can cause severe health hazards and are listed as priority pollutants by many environmental protection agencies worldwide (Jaishankar et al., 2014; Dixit et al., 2015; Sarwar et al., 2017). Therefore, the removal of HMs from the contaminated matrix is an urgent need to safeguard the environment and human health. Phytoremediation has been identified as an emerging, low-cost and eco-sustainable solution for HM pollution prevention and control. It is the most suitable alternative to conventional physicochemical remediation technologies, which are highly expensive, technically more suited to small areas, create secondary pollution and deteriorate soil fertility and thus, adversely affects agro-ecosystem (Ali et al. 2013; Chandra et al. 2015; Mahar et al. 2016; Muthusaravanan et al. 2018).

Phytoremediation is the engineered use of green plants with associated soil beneficial microbes to remove toxic pollutants *via* degradation and detoxification mechanisms from contaminated soil and water/wastewaters (Bharagava et al. 2017a; Mukhopadhyay and Maiti 2010; Ali et al. 2013). It is an eco-

friendly, non-intrusive and aesthetically pleasing remediation technology that removes metal pollutants from the contaminated sites (Lee 2013; Chandra et al. 2015; Chirakkara et al. 2016). It can be commercialized and income can be generated, if metals removed from contaminated sites could be used to extract usable form of economically viable metals (i.e. phytomining) (Chandra et al. 2015; Mahar et al. 2016). In addition, energy can be generated through the burning of plant biomass and land restoration could be achieved for sustainable agricultural development or general habitation (Stephenson and Black 2014; Mahar et al. 2016). The rationale, mechanisms and economic feasibility of phytoremediation have been discussed elsewhere (Ali et al. 2013; Wan et al. 2016; Sarwar et al. 2017). However, extensive research is currently underway to testify the phytoremediation potential of hyperaccumulating plants at field scale for the treatment and management of HM-contaminated sites.

The deadly poisonous and indestructible nature of HMs is mainly responsible for the eco-toxicity and health hazards. Thus, an eco-friendly solution (i.e. phytoremediation) is required for the treatment and management of HMcontaminated sites. This paper aims to provide a comprehensive review on the following areas of phytoremediation: (a) environmental pollution and toxicity profile of HMs; (b) conventional and novel phytoremediation approaches and their role in environmental management with merits and demerits; (c) field studies and sustainability issues associated with phytoremediation of HMs-contaminated sites; (d) plant-microbe interactions (PMIs) and their role in enhanced phytoremediation; (e) challenges and opportunities for valorization of plant biomass in biofuel/bioenergy production; (f) challenges in transgenic approaches to modify the hyperaccumulating plants (designer plants) and associated microbes (engineered bacteria); and (g) the knowledge gaps and potential areas for further research in the phytoremediation of HMs-contaminated sites.

2   Sources of Heavy Metals Contamination and Toxicity in Environment

HMs can be introduced into the environment either by natural or anthropogenic processes. Natural processes are geological activities; for instance, mineral weathering, erosion, volcanic eruptions, and continental dust. Anthropogenic activities include industrial operations such as mining, smelting, electroplating, and

industrial effluent discharge as well as agricultural practices like use of pesticides and phosphate fertilizers and release of agricultural wastes (Ali et al. 2013; Mahar et al. 2016; Antoniadis et al. 2017). Industrial activities are the major source of HM pollution (water and soil) in the environment. If HMs enters the food chain, they may bioaccumulate and/or biomagnify at higher trophic levels resulting in severe health threats and thus, are of serious eco-toxicological concern.

The indiscriminate discharge of toxic metals-rich industrial effluents is one of the major sources of environmental pollution. The effluent discharged from metal-based industries, especially leather industries (Cr used in leather tanning) cause serious soil and water pollution and hence, its treatment and management is a key challenge to pollution control authorities (Sahu et al. 2007; Saxena et al. 2016). A high concentration of HMs has been reported in sediments of river Ganga and its tributaries receiving Cr-loaded tannery effluent (Beg and Ali 2008). In addition, HMs beyond the permissible limits also deteriorates water quality and makes it unfit for drinking and irrigation purpose (Nazeer et al. 2014). The effluent released from electroplating and distillery industries also constitute a highly rich source of HMs and hence, considered as hazardous to living beings (Venkateswaran et al. 2007; Chandra et al. 2008). Furthermore, effluent released from domestic activities is also responsible for HM pollution and thus, is of serious eco-toxicological concerns (Bhardwaj et al. 2017).

In an aquatic ecosystem, HMs adversely affects gamete production, sperm quality, embryonic development, delay hatching and causes physical deformities in fishes and ultimately, leads to the death of newly hatched larvae (Segura et al. 2006; Jezierska et al. 2009; Fatima et al. 2014). HMs also causes endocrine disruption, oxidative stress and genotoxicity in fishes (Jezierska et al. 2009; Luszczek-Trojnar et al. 2014; Javed et al. 2016). Further, HMs also causes a reduction in hematological parameters and glycogen reserve and thus, make the fishes weak, anemic and vulnerable to diseases (Javed and Usmani 2015).

The soil is a non-renewable resource for sustainable agriculture and acts as a major sink for HMs. The contamination of agricultural soil with toxic metals affects its physico-chemical and biological properties and reduces land usability for agricultural farming leading to food insecurity and thus, creates land tenure

problems (Wuana and Okieimen 2011). Moreover, the co-existence and persistence of HMs in soil is also responsible for the entry of toxic metals into the food chain and thus, lead to severe health hazards in living beings (Khan et al. 2008) (Table 1).

HMs inhibits several microbial metabolic processes such as respiration, denitrification, and enzymatic activity and hence, retard the bioremediation processes (Zhuang et al. 2007; Sobolev and Begonia 2008). HMs also causes a reduction in the number of specific microbial populations and a shift in the microbial community structure. For instance, Ding et al. (2017) evaluated the effect of Cd and Cr on the microbial community structure in the rhizospheric soil of rice plant during a pot experiment. Results revealed that the relative abundance of a bacterial genus, Longilinea was significantly higher in the control soil than in Cd and Cr-treated soils whereas the relative abundance of the genus, Pseudomonas was significantly higher in the Cd-treated soils than in the Cr-treated and control soils. However, the relative abundance of a genus, Sulfuricurvum was also significantly higher in the Cdtreated soil than in the Cr-treated and control soils whereas the relative abundance of the genus, Bellilinea was significantly higher in the Cr-treated soil than in the other treated soils. HMs also inhibit the cell division, transcription process, denaturation of protein and adversely affect the cell membrane distribution in microbes (Jacob et al. 2018). Hexavalent chromium (Cr^{6+}) is also reported to cause DNA damage by exerting oxidative stress in soil bacteria and thus, leads to genotoxic effects (Quievryn et al. 2003).

The irrigation of food crops in the agriculture field with water contaminated with toxic metals-rich industrial effluents is a common practice in many developing countries. It may provide a chance for the movement of potentially toxic metals from contaminated soil to edible crops, ultimately, reaches into the human/animal body via consumption, and thus, renders severe toxic effects. HMs affects various metal-sensitive enzymes in plants such as alcohol dehydrogenase, nitrogenase, nitrate reductase, amylase, and hydrolytic (phosphatase and ribonuclease), and carboxylating (phosphoenolpyruvatecarboxylase and ribulose-1,5-bisphosphate carboxylase) enzymes (Nagajyoti et al. 2010; Yadav 2010). Hence, HMs disrupts several biochemical/physiological processes in plants such as seed germination, enzymatic activities,

nitrogen metabolism, electron transport system, transpiration, CO_2 assimilation, antioxidant defense system, photosynthesis, photophosphorylation, cellular metabolism, nitrogen fixation, water balance, mineral nutrition, cellular ionic homeostasis and ultimately, leads to plants death (Yadav 2010; Lajayar et al. 2017). Irrigation of agricultural crops with heavy metal-loaded industrial effluents also disrupt several cytological processes in plants such as root growth and elongation, cell membrane permeability, mitotic activity, the stability of genetic material and also create chromosomal abnormalities (Nagajyoti et al. 2010; Yadav 2010). For example, the irrigation of agricultural crops with the HM-rich distillery and tannery effluent have been reported to cause a reduction in root/shoot growth and biomass, seed germination, seedling growth, and also induce chlorosis, photosynthetic impairment (Chandra et al. 2009; Bharagava et al. 2017b).

HMs may cause oxidative stress by forming reactive oxygen species (ROS), which disrupt the antioxidants defense system and lead to cell damage in humans/animals; and in extreme cases can be fatal (Jaishankar et al. 2014). For instance, hexavalent chromium (Cr^{6+}) has been reported to cause cancer in humans and damage cellular components during its reduction into trivalent chromium (Cr^{3+}), leading to the generation of free radicals that cause DNA damage (Mishra and Bharagava 2016). Therefore, the remediation of HM-contaminated sites is utmost important for environmental safety.

3   Trophic Transfer of Toxic Heavy Metals and Its Consequences

Trophic transfer or biotransference is an ecological phenomenon by which a contaminant enters the food chain through uptake either from ambient abiotic environment (bioconcentration) or both ambient abiotic environment and organism's food/diet (bioaccumulation), passage from one trophic level to the next higher trophic level (biomagnification) and consequently poses risks to human/animal health (Ali and Khan 2018). The trophic transfer of toxic HMs from soil to plants to humans and organism's food to humans is depicted in Fig. 1. The primary route of HM entry into the food chain is through the soil-to-plant transfer mechanism. In the soil-to-plant transfer mechanism, HMs are transferred from soil to agricultural crops/vegetables that constitute a large source of human diet and thus, may result in catastrophic health hazards (Table 1). According to a study, the daily intake of metal (DIM) were higher for vegetables grown on soils irrigated

with HMs rich-wastewater compared to those of control soils (Jan et al. 2010). The consumption of fishes contaminated with toxic HMs also poses serious risks to human health. Hence, their dietary role has been questioned though they provide omega-3 fatty acids that have cardioprotective effects. In the 1950s, Minamata disease in Japan caused by the consumption of Hg-contaminated fishes by the local people is considered as one of the major environmental chemical disasters of the 20th century (Ali and Khan 2017). Moreover, HMs accumulation in invertebrates (due to varying feeding habits of insects), amphibians and reptiles (due to absorption through highly permeable skin), and birds (due to ingestion of contaminated food and water) also adversely affect their development, growth, health, feeding behavior, physiology, and reproduction (Ali and Khan 2018b). Thus, the trophic transfer, bioaccumulation, and biomagnification of toxic HMs in food chains have important implications for wildlife and human health. Further, more details on the subject can be found in a good review article published by Ali and Khan (2018).

4 Phytoremediation Approaches for Environmental Cleanup

The engineered use of green plants with associated beneficial microbes to degrade/detoxify pollutants from the contaminated medium (soil/water/wastewaters) is technically described as phytoremediation. The term "phytoremediation: is made up of two words i.e. Greek head *"phyto"* (means plant) and Latin root *"remedium"* (means to correct or remove an evil). It can be applied for the eco-restoration of sites primarily contaminated with HMs, radionuclides and various recalcitrant organic pollutants (Ali et al. 2013; Mahar et al. 2016). It comprises different phytotechniques for the amelioration of various pollutants using different mechanisms depending on their applications (Fig. 2). However, all the mechanisms cannot be applied for the remedy of all the pollutants. Different phytoremediation techniques such as phytoextraction, phytostabilization, phytovolatilization, rhizodegradation, phytodegradation, and rhizofiltration have been extensively discussed elsewhere (Ali et al. 2013; Chandra et al. 2015; Chirakkara et al. 2016; Sarwar et al. 2017). The definition, application, and bottlenecks of traditional phytoremediation techniques are summarized in Table 2.

Among the phytoremediation techniques, phytoextraction is a major mechanism of HM removal from contaminated sites. When green plants are used to remove metal pollutants from contaminated sites *via* root absorption and their sequestration/concentration in the above-ground harvestable parts is technically described as phytoextraction (Vangronsveld et al. 2009; Mukhopadhyay and Maiti 2010; Mahar et al. 2016). It could be an economically viable technology when metals extracted from contaminated sites using plants biomass can be utilized as "bio-ore" to extract the functional and valuable metals and this process is technically regarded as phytomining

(Chandra et al. 2015). Thus, it can generate income and offer additional employment opportunity to the public (Sheoran et al. 2009; Chandra et al. 2015; Stephenson and Black 2014).

The phytoextraction efficiency of green plants primarily depends on the bioconcentration factor (BCF) and translocation factor (TF). BCF represents metal concentration in root/soil and denotes metal accumulation whereas TF represents metal concentration in shoot/root and denotes metal translocation (Goel et al. 2009; Ali et al. 2013; Antoniadis et al. 2017). Plants with high biomass, fast growth rate, and high metals tolerance and accumulation are chiefly preferred for metal's phytoextraction (Mukhopadhyay and Maiti 2010; Lee 2013; Chandra et al. 2015). Phytoextraction is performed in two different ways (Ali et al. 2013): natural (plants accumulate metals under natural conditions) and induced or assisted (application of enhancers to increase metal accumulation in plants). Enhancers are used to increase the phytoextraction efficiency and include chelators or soil amendments (Sarwar et al. 2017). Chelators are the organic and mineral acids that increase the bioavailability of insoluble or unavailable form of metals in soil and makes them available for plants uptake (Ali et al. 2013; Mahar et al. 2016). Thus, enhance the phytoremediation efficiency by solving the low metal phytoavailability issue. Some commonly available chelators are EDTA: ethylenediaminetetraacetic acid; HEDTA: N-hydroxyethylenediaminetriacetic acid; DTPA: diethylenetriaminepentaaceticacid; EGTA: ethyleneglycolbis(b-aminoethyl ether),N,N,N0 ,N-tetraacetic acid; EDDHA: ethylenediamine-di(o-hydroxyphenylacetic acid); EDDS: ethylenediamine-N,N'-disuccinic acid; NTA: nitrilotriacetic acid, and CA: citric acid. The ability of chelators to enhance the metal

accumulation in plants has been tested and reviewed by many workers (Xie et al. 2012; Ramamurthy and Memarian 2014; Sun et al. 2015; Chirakkara et al. 2016). Organic soil amendments are cheaper, eco-friendly, and non- or less toxic and degradable in nature. These help to minimize environmental pollution and reduce toxicity to remediating plants and ultimately, enhance phytoremediation efficiency (Wiszniewska et al. 2016). Some specific kinds of organic soil amendments include agro- and industrial wastes (such as sugar beet residue, wheat, and rice straw, composted sewage sludge or molasses), biochar, compost, humic substances, plant extracts, and exudates and are of great significance in HMphytoremediation (Wiszniewska et al. 2016). The use of organic soil amendments to enhance the phytoremediation efficiency has been evaluated (Park et al. 2011; Paz-Ferreiro et al. 2014; Wiszniewska et al. 2016; Chirakkara et al.

2016; Reddy et al. 2017). The biotic and abiotic factors are also affect the efficiency of phytoremediation (Fig. 3). Biotic factors include plant and root zone characteristics whereas abiotic factors comprise pollutants and chelators characteristics, properties of the medium (e.g. soil) and climate conditions.

Further, the environmental risks associated with synthetic chelators such as low biodegradability of chelators, groundwater contamination due to leaching of highly water-soluble metal-chelator complexes to deeper layer of soil, slow decomposition of organic acids, and toxicity to remediating plants and soil beneficial microorganisms (SBMs) should also be considered before application (Vangronsveld et al. 2009; Stephenson and Black 2014; Mahar et al. 2016). However, the selection of chelators with optimum dose and application time could help to minimize the associated environmental risks and toxic effects in remediating plants.

5   Hyperaccumulating Plants for Phytoremediation of Heavy Metals

5.1   *Classification of metallophytes*

Plants that tolerate and survive in soil containing an exceptionally high concentration of HMs without suffering toxicity are termed as metallophytes. The soil, where ores are outcropping is termed as metalliferous or orogenic soil. According to growth potential in HMs-contaminated sites, metallophytes can

be grouped into the following categories: (a) metal excluders (those accumulate metals in their roots but restrict transport and entry into their aerial parts possibly by altering cell membrane permeability and changing cell wall metal binding capacity *via* modulating ionic channels, ion pump activity, and activation of new ionic conductance or exudating more chelating substances in soil); (b) metal indicators (those actively accumulate metals in their aerial parts by releasing intracellular metal binding chemicals i.e. chelators, or altering the pattern of metal compartmentalization by storing them in nonsensitive plant parts such as vacuoles and cell wall and generally reflect metal concentration in soil); and (c) metal accumulators (those actively accumulate exceedingly large concentration of metals from the soil in the aboveground plant parts, especially leaves with no symptoms of phytotoxicity) (Mukhopadhyay and Maiti 2010; Chandra et al. 2015; Antoniadis et al. 2017). The use of metallophytes alone or in combination with microorganisms is an excellent strategy for the phytoremediation and HM pollution prevention and control.

To date, several metallophytes have been identified and used in the phytoremediation of HMs-contaminated sites. Some specific examples include *Pteris vittata,* which can accumulate Cr and As up to 35,303 and 20,707 mg kg^{-1} dry weight (DW), respectively (Kalve et al. 2011), *Alyssum murale* can accumulate Ni in range of 4730-20100 mg kg^{-1} DW (Bani et al. 2010), *Tagetes minuta* can accumulate As up to 380.5 mg kg^{-1} DW (Salazar and Pignata 2014), *Eleocharis acicularis* can accumulate Zn up to 11,200 mg kg^{-1} DW (Sakakibara et al. 2011), *Corrigiola telephiifolia* can accumulate As up to 2110 mg kg^{-1} DW (Garcia-Salgado et al. 2012), and *Noccaea caerulescens* can accumulate Pb in range of 1700-2300 mg kg^{-1} DW (Dinh et al. 2018).

5.2 *Selection Criteria for Hyperaccumulating Plants for Phytoremediation*

Plants (woody/herbaceous) that accumulate high metals concentration in their shoot (100-1000 fold higher than those found in non-hyperaccumulating species) without any visible symptoms are regarded as hyperaccumulators (represented by < 0.2% of angiosperms) and overall process is termed as hyperaccumulation (Ghosh and Singh 2005; Mukhopadhyay and Maiti 2010; Lee 2013). An ideal hyperaccumulator plant should accumulate at least 100 mg kg^{-1} (0.01% dry wt.) of As and Cd, 1000 mg kg^{-}

[1] (0.1 % dry wt.) of Co, Cu, Cr, Ni, Se and Pb, and 10,000 mg kg^{-1} (1% dry wt.) of Zn and Mn (Reeves and Baker 2000). Metal hyperaccumulating plants are chiefly preferred for phytoremediation. However, some hyperaccumulating plants are not successfully applied because of several reasons: (a) low biomass, (b) slow growth rate, (c) metals (such as Ni, Zn, and Cu) under scope of phytoextraction are not the priority pollutants, and (d) agronomic practices and crop protection measures for their cultivation and protection have not been developed (Goel et al. 2009; Marques et al. 2009; Mahar et al. 2016; Yadav et al. 2018).

Serpentine/ultramafic soils (most abundant metalliferous soil on earth) are the most important natural resource in the screening of hyperaccumulating plants especially for metals like Ni and/or Zn. Plant species inhabiting such soils are called as "serpentinophytes". Serpentinophytes are of great significance in phytoremediation due to their specific adaptation (might be confined to the physiological state) to such high HMs-rich soil. Several plant species like *Crotalaria micans*, *Leucaena leucocephala*, *Bidens pilosa*, *Pueraria lobata*, and *Conyza canadensis* have been reported to remove Ni from a serpentine site for successful phytoremediation applications in Taiwan (Ho et al. 2013). Similarly, many research reports have been published on the screening of hyperaccumulating plants for metals like Zn and Ni from serpentine soils (Ho et al. 2013; Tomovic et al. 2013; Salihaj et al. 2016; Bini et al. 2017). According to Bini et al. (2017), *Alyssum bertolonii* accumulated an exceptionally high concentration of Ni

(i.e. 2118 mg kg^{-1} in aerial parts) from serpentine soils of Tuscany (Central Italy). They also suggested the use of *Alyssum bertoloni* in the remediation of Ni-contaminated soils with both the phytoextraction and phytostabilization and in phytomining as well. Thus, serpentinophytes are the key metal hyperaccumulators and amicable for the phytoremediation and phytomining. However, the ecological significance of metal hyperaccumulation in plants is still unclear. It has been suggested that the hyperaccumulation in plants might result from the development of high metal tolerance, drought resistance, unintended metals absorption, competition with other metal-tolerant plants, and defense mechanisms for the protection against herbivores and pathogens (Lee 2013; Mahar et al. 2016). Further, unveiling the ecological roles of metal hyperaccumulation in plants may help us to clearly understand the mechanism of phytoremediation.

The selection of hyperaccumulating plants for phytoremediation purpose is a tedious task. Hence, indigenous plants are often recommended due to their high metal removal efficiency, less competition among them under local environmental conditions, and the lesser threat of becoming invasive (Ghosh and Singh 2005; Mukhopadhyay and Maiti 2010; Chandra et al. 2015). The ideal plants for phytoremediation should possess a series of characteristics: (a) ability to hyperaccumulate HMs preferably in the aboveground parts; (b) tolerance to high pH, salt and accumulated toxic metals concentration; (c) fast growth and high biomass; (d) widespread highly branched roots; (e) easy to cultivable and harvestable; and (f) resistant to diseases and pests (Vangronsveld et al. 2009; Chandra et al. 2015; Mahar et al. 2016). To date, some hyperaccumulator satisfies the above said criteria to be applied for HMs phytoremediation. Metal hyperaccumulating plants are often preferred over non-accumulators because they produce a high volume of metal-rich biomass and are economic to process for metal recovery and safe disposal, which have an additional eco-environmental benefit. To date, more than 500 plant species have been identified as metalhyperaccumulators. Notable representative includes the members of *Brassicaceae, Asteraceae, Caryophyllaceae, Lamiaceae, Euphorbiaceae, Poaceae* etc., and have been reviewed elsewhere (Ali et al. 2013; Chandra et al. 2015; Mahar et al. 2016). Some examples of hyperaccumulating plants with their metal accumulation capacity are listed in

Table 3.

6   Emerging Halophytes in Phytoremediation

Plants that tolerate and thrive in a highly saline environment with an extreme salt concentration (200 mM NaCl or more) are termed as halophytes (Liang et al. 2017). Halophytes are cultivable in soils irrigated with highly saline water/wastewater where good quality water may not be available or limited in supply (due to high urban requirements and climate change) for crops irrigation in the arid environment (Manousaki and Kalogerakis 2011). It represents an additional eco-environmental benefit for phytoremediation. High salinity increases the mobility of HMs in soil and therefore, facilitates their greater uptake and translocation

from root to shoot to achieve phytoremediation (Wang et al. 2014; Liang et al. 2017). For example, the halophyte, *Tamarix smyrnensis* removes

9.4, 19.7 and 38.3 μg of Cd in a solution containing 0, 0.5 and 3% NaCl, respectively (Manousaki et al. 2008).

High salt (like NaCl) concentration increase HM accumulation in halophytes either by enhancing metal mobility or by modifying root functions and alleviate metals-induced phytotoxicity through improved management of osmotic solutes and oxidative status (Chai et al. 2013). Thus, halophytes are suitable candidates for the phytoremediation of HM-contaminated saline soils (Table 4). The role of salinity in increasing Cd bioavailability is mainly attributable to the formation of Cd-Cl complexes ($CdCl^+$ and $CdCl_2$) (Weggler-Beaton et al. 2000; Chai et al. 2013). These complexes are less strongly sorbed to soil than a free Cd^{2+} ion and hence, increase Cd mobility at the soil-root interface. Moreover, these complexes also stimulate transport of Cd across the zone encompassing soilrhizosphere apoplast-plasma membrane. Thus, increased soil-plant transfer of Cd can occur under salinity. Although the detailed interaction between salinity and HMs accumulation is still not fully understood.

The halophytes applied for the phytoremediation of HM-contaminated saline soils have been recently reviewed (Oosten and Maggio 2015; Liang et al. 2017). The mechanism of metals and salt tolerance in halophytes include; (a) osmotic adjustment through ion accumulation/compartmentalization or exclusion, and biosynthesis of compatible solutes; (b) involvement of antioxidant defense system; (c) cell walls and sub-cellular compartmentalization; (d) metal chelation or detoxification; and (e) metal excretion and complexing ligands (Liang et al. 2017). Recently, "phytoexcretion" has been introduced as a novel phytoremediation process for salt affected metal-contaminated sites (Liang et al. 2017). It is a type of metal detoxification strategy in halophytes, wherein toxic metals are excreted through specialized salt glands from leaf tissue onto leaf surface (Manousaki and Kalogerakis 2011; Liang et al. 2017). Thus, applying phytoextraction in conjunction with phytoexcretion using halophytes represents a promising strategy for the phytoremediation of high salt affected metal-contaminated sites.

Halophytes such as *Atriplex halimus*, *Echinochloa stagnina*, *Spartina alterniflora*, *Zygophyllum fabago*, *Sesuvium portulacastrum,* and *Tamarix africana* have been well reported for the phytoextraction/phytostabilization of HM-contaminated saline soils (Liang et al. 2017). The use of halophyte, *Atriplex nummularia* in the remediation of saline and sodic soils is also reported due to its high biomass and salt extraction capability (de Souza et al. 2014).

However, the suitability of different halophytes for the phytoremediation of HM-contaminated sites is still under evaluation. The future research should be focused on the: (a) mechanistic understanding of simultaneous salt and HM tolerance; (b) use of halophytes of economic importance for HM phytoremediation to gain better economic returns; (c) use of transgenics to develop engineered halophytes with high biomass and fast growth rate for effective HM phytoremediation under abiotic stress; and (d) use of biochar and other soil amendments for the improved phytoremediation of salt-affected HM-contaminated sites (Liang et al. 2017).

7   Medicinal and Aromatic Plants in Phytoremediation

Generally, edible crops are not suitable for the phytoremediation of HMs-contaminated sites due to the risk of entering into the food chain *via* consumption by humans/animals and associated health hazards. The application of medicinal and aromatic plants for the phytoremediation of HM-contaminated sites could be an innovative approach. Aromatic plants are mostly non-edible and are not being consumed directly by humans or animal due to their essence. They have low metal accumulation potential as compared to edible plant hyperaccumulators but are economically important as their harvested foliage are the chief source of essential oil. The essential oil obtained from aromatic plants is free from the risk of HMs accumulation from plant biomass and thus, prevents entry of HMs into the food chain (Gupta et al. 2013; Lajayar et al. 2017). HMs remains in the extracted plant residues during oil extraction through the distillation process and hence, limits the detectable concentration in the essential oil. The export of essential oil for selling is a major economic incentive along with phytoremediation because it is being used in soaps, detergents, insect repellents, cosmetic, perfumes, and food processing industries (Gupta et al. 2013; Lajayar et al. 2017).

Some aromatic plants, such as *Artemisia annua, Mentha arvensis, Cannabis sativa, Lavandula vera, Matricaria chamomilla, Mellissa officinalis, Ocimum gratissimum, Portulaca oleracea;* and *Salvia officinalis* have been investigated for the phytoremediation of HM-contaminated soil (Lajayar et al. 2017) (Table 4). However, due to higher ability of some aromatic plants to accumulate HMs in their aerial parts, when the intention of their cultivation on contaminated soil is non-phytoremediation (i.e. edible consumption), the consumption of such aromatic plants may result in serious health hazards and thus, require continuous monitoring (Lajayar et al. 2017). The phytoremediation of HM-contaminated sites using aromatic plants is a newly emerging concept and currently under research. There are very few available studies to date and hence, additional research is required to explore their potential in phytoremediation. Furthermore, transgenic approaches could be used to enhance the metal accumulation capacity in such plants. For instance, Dhankher et al. (2002) developed transgenic *Arabidopsis thaliana* plants (medicinal herb used to cure sores in the mouth) with increased tolerance and accumulation of As for enhanced phytoremediation.

8   Molecular Mechanism of Heavy Metals Tolerance, Uptake, Translocation, and phytoremediation

HMs are highly toxic to plants as they disturb the redox status (balance between oxidants and antioxidants) and lead to oxidative stress responsible for the physiological damage. Plants always maintain a very low concentration of free radicals to avoid any physiological damage. This balance is established by the uptake and translocation of toxic metals, their sequestration, and binding to proteins and organic ligands. Plants uptake toxic metals from polluted matrix *via* roots and store them either in roots or translocate to the shoots through xylem vessels, where they sequester in the vacuoles. Vacuoles help to reduce the excess metal ions from the cytosol and, prevent their interactions with other metabolic processes due to low metabolic activity and thus, are considered to be the ideal sites of metals sequestration (Wu et al. 2010; Ali et al. 2013; Chandra et al. 2015). Metal tolerance is a pivotal requirement for metal accumulation and phytoremediation and governed by a variety of biomolecules. Membrane transporter proteins (MTPs) such as ATPases, zinc-iron permease (ZIP), cation diffusion facilitators (CDF), copper transporters (COPTs), ATP-binding cassettes

(ABC), and cation exchangers (CAXs) and heavy metal ATPase (HMA) such as P1B-ATPase, HMA4 and HMA5 help in the uptake and transport of metals across the cell membrane and facilitate their detoxification (Ali et al. 2013; Sarwar et al. 2017). Metal binding proteins (such as Cu-chaperone ATX1-like proteins, glutathione (GSH), metallothioneins (MTs) and phytochelatins (PCs)) and organic ligands are involved in the binding, sequestration and detoxification of toxic metals in the above-ground plant parts preferably in the cuticle, epidermis and trichomes as these have less chance of cellular damage (Wu et al. 2010; Chandra et al. 2015; Sarwar et al. 2017). GSH protect the plants from physiological damage caused by toxic metal stress, MTs reduce metal accumulation in shoots by trapping them in roots and PCs enhance HM-tolerance, accumulation, and detoxification in plants (Goel et al. 2009; Wu et al. 2010). Thiol groups present in these biomolecules form complexes with HMs (GSH-PC-MT-HM) and thus, play a crucial role in the detoxification of toxic metals in plants (Sarwar et al. 2017).

9   Exploiting Plant-Microbe Interactions for Enhanced Metals Phytoremediation

The longtime frame required for phytoremediation and physiological damage to remediating plants under toxic metal stress is a major issue. Therefore, exploiting plant-microbe interactions (PMIs) could be exploited to enhance the plant growth and phytoremediation of HMs-contaminated sites.

The root/rhizosphere colonizing, plant growth promoting rhizobacteria (PGPR) have been reported to enhance host plant growth in toxic metals-contaminated sites (Yuan et al. 2013; Ma et al. 2015; 2016a). PGPR produces growth hormones such as auxins (IAA: Indole-3-acetic acid), cytokinins, gibberellins and ethylene (Rajkumar et al. 2012; Ma et al. 2015). The mechanisms of plant growth promotion may vary from bacterial strain to strain and depends on various secondary metabolites produced (Ma et al. 2011; Backer et al. 2018). PGPR also produces some other beneficial compounds such as enzymes, osmolytes, biosurfactants, organic acids, metal chelating siderophores, nitric oxide, and antibiotics (Rajkumar et al. 2012; Ma et al. 2015). These beneficial compounds reduces ethylene production *via* synthesis of ACC (1-aminocyclopropane-1-carboxylate) deaminase that prevents the inhibition of root elongation, lateral root growth, and root hair formation, and also improve the minerals (N, P & K) uptake in acidic soil (Babu et al.

2013; Ma et al. 2015). These compounds also suppress phytopathogens, provide tolerance to abiotic stress and helps in associated nitrogen fixation (Rajkumar et al. 2012; Babu et al. 2013; Ma et al. 2015). Hence, PGPR are applied in sustainable agriculture development. Besides these, PGPR can lower the metal toxicity to remediating plants through biosorption/bioaccumulation as bacterial cells have an extremely high ratio of surface area to volume (Ma et al. 2016b; Li et al. 2018). PGPR could adsorb high metals concentration by either a metabolism-independent passive or metabolism-dependent active processes. Hence, using PGPR in environmental bioremediation could be a useful strategy for plants survival in the stressed environment. PGPR reported for the enhanced HMs phytoremediation with associated benefits have been reviewed in past (Ma et al. 2011; Rajkumar et al. 2012; Ullah et al. 2015). Some updated examples from recent studies are summarized in Table 5.

Endophytes are the microbes (bacteria/fungi) that reside in the inner tissues of plants without causing harm to host. They also help in plant growth promotion and development under biotic or abiotic stressed environment and exert many beneficial effects than rhizobacteria (Luo et al. 2011; Ma et al. 2011; 2015). They are able to tolerate high metals concentration and hence, lower phytotoxicity to remediating plants as well as helps in growth promotion enhancing through biocontrol mechanism and induced systemic resistance against phytopathogens (Ma et al. 2011; 2015). They produce phytohormones, organic acids, siderophores, biosurfactants, enzymes, and growth regulators that help in water and nutrients (P, N & K) uptake, osmolytes accumulation, osmotic adjustment, stomatal regulation and associated nitrogen fixation as additional benefits to host plants (Ma et al. 2011; 2016b). Thus, inoculating plants with endophytes could be an excellent strategy to enhance the phytoremediation of HM-contaminated sites. Endophytes applied to enhance HMs phytoremediation with associated benefits have been recently reviewed by several researchers (Afzal et al. 2014; Ma et al. 2016b).

Arbuscular mycorrhizal fungi (AMF: colonize plant roots) have been also reported to protect their host plants against heavy metal toxicity through their mobilization from soil and thus, helps in phytoremediation (Marques et al. 2009; Meier et al. 2012; Khan et al. 2014). The possible mechanisms by which AMF protect

their host plants through metal mobilization from soil include: (a) immobilization by chelation; (b) binding of metals to biopolymers in the cell wall; (c) superficial immobilization in the plasmatic membrane once metals crosses the cell wall; (d) membrane transportation that mobilizes metals from the soil to the cytosol (e) intracellular chelation through MTs, organic acids, and amino acids; (f) export of metals from cytosol by membrane transporters; (g) sequestration of metals into vacuoles; (h) transportation of metals by means of fungal hyphae; (i) storage of metals in fungal spores; and (j) exportation by the fungus, and access into the plant cells, involving both active and passive transportation into the mycorrhizae (Meier et al. 2012; Cabral et al. 2015). They confer resistance against drought, high salt, and toxic metals concentration, and improve nutrient supply and soil physical properties (Khan et al. 2014). The exact mechanism of plant protection is still not fully understood and further research is required to explore their role in the phytoremediation. In addition, isolating and characterizing suitable plant associated beneficial microbes is a timeconsuming process. It also requires the analysis of more than thousands of isolates and thus, identification of specific biomarkers may help to select the effective plant-microbe associationships for microbe-assisted phytoremediation (Rajkumar et al. 2012). Further, to ameliorate metal toxicity, plant growth promotion and metal sequestration, extensive research efforts are also required to explore novel microbial diversity, their distribution, as well as functions in the autochthonous and allochthonous soil habitats for microbe-assisted phytoremediation of

HM-contaminated sites.

10   Molecular Approaches for Enhanced Phytoremediation of Heavy Metals

Transgenic approaches are decisive in genetic manipulation of low biomass and slow-growing hyperaccumulating plants to enhance the phytoremediation of HM-contaminated sites. The main objective is to introduce genes (from organisms such as plants, bacteria, and mammals) that confer plants the ability to resist, tolerate and hyperaccumulate toxic metals from contaminated sites under changeable environment with increased biomass and metal storage capacity (Goel et al. 2009; Marques et al. 2009; Mukhopadhyay and Maiti 2010). The general approach behind the transgenic approaches is to over-express or knockdown

genes that encode for metal binding PCs, metal chelating MTs and MTPs, which are crucial in toxic metals detoxification and thus, phytoremediation (Goel et al. 2009; Mukhopadhyay and Maiti 2010; Dhankher et al. 2011; Sarwar et al. 2017).

Chloroplast engineering is an innovative approach that allows the transfer of entire operons from bacteria into plants for the overexpression of enzymes responsible for HM phytoremediation (Dhankher et al. 2011; Goel et al. 2013). Identification of novel genes and their transfer from natural hyperaccumulators and microbes into fastgrowing metal hyperaccumulators may also create new opportunities for enhancing phytoremediation efficiency. To date, transgenic plants have not yet been applied for field scale application and genes escaping from transgenic plants to wild relatives are very rare. Genetically engineered plants (GEPs) applied for the enhanced metals phytoremediation in the laboratory has been reviewed (Kotrba et al. 2009; Goel et al. 2009; Vangronsveld et al. 2009; Chandra et al. 2015). Some updated examples from the recent studies are summarized in Table 6. However, there are several environmental risks associated with transgenic plants applied for metals phytoremediation at the field should also be considered. Environmental risks may include (a) exposure of toxic metals to wildlife and humans to their more bioavailable forms; (b) uncontrolled spread of transgenic plants due to interbreeding with their wild relatives or superior fitness because of weedy nature in environment (genetic pollution); (c) transformation of natural flora through cross-pollination; and (d) risk of invasion of free plants and potential loss of diversity (Kotrba et al. 2009; Marques et al. 2009; Wu et al. 2010). Hence, risk assessment should be performed before applying GEPs at the field scale.

The engineering of symbiotic plant-associated microbes (i.e. PGPRs and endophytes) could also be a promising phytobacterial technology to enhance the tolerance to high metal concentrations and detoxification (Weyens et al. 2013; Huang et al. 2016). It involves the introduction of one or more genes of interest that code for enzymes responsible for the enhanced remediation, stress tolerance, metals chelators, uptake regulators, transporters and homeostasis (Ullah et al. 2015). Some updated examples from the recent studies on the genetically engineered bacteria applied for enhanced metals phytoremediation are listed in

Table 7. However, engineering plant-associated microbe are of limited scope as these mainly concentrate around the roots, show limited distribution outside rhizosphere and depends chiefly on the host plants (Wu et al. 2006). Hence, their controlled use outside rhizosphere is not an easy task

Recent advances in "omics" technologies (such as proteomics, metabolomics, genomics, metagenomics, and transcriptomics) also offer greater opportunities to identify traits that maximize the benefits of phytoremediation through manipulating tolerance, accumulation, and pollutants degradation/detoxification potential of plants and microbes. Thus, the insertion and overexpression of genes and metal-binding proteins and their exploitation to increase metal-binding capacity and tolerance or accumulation of toxic metals in bacteria and plants could be an excellent strategy for the enhanced phytoremediation.

11   Energy Crops in Phytoremediation and Bioenergy Production

The treatment and safe disposal of huge quantity of metals-contaminated biomass associated with phytoextraction is an environmental concern. Non-edible and perennial energy crops can be used to maximize the benefits of phytoremediation. Energy crops have high density, biomass, and mechanization, fast growth rate, and short rotation time and are resistant to diseases and pests. The biomass of energy crops could be economically valorized for renewable energy (biogas, biofuels, and combustion for energy generation and heating) production to fulfill the global energy demands, which is one of the major challenges of the 21st century (Lavanya et al. 2012; Ahmad et al. 2016). An energy crop, *Miscanthus x giganteus* has been predicted to supply up to 12% energy of the total energy need of the European Union (EU) (Fruhwirth and Liebhard 2004). Applying energy crops in phytoremediation represents as an additional eco-environmental benefit in controlling soil erosion, improving soil health and providing the wildlife habitat (Simpson et al. 2009; Gomes 2012). It could also be an economic incentive for phytoremediation, particularly when energy crops are grown in barren metal-contaminated land that does not compete for food production. It will reduce the consumption of non-renewable fossil fuels and creates wide employment opportunities for locals with the low-impact treatment of barren metals-contaminated

lands (Gomes 2012). A list of energy crops used for the phytoremediation and biofuels/bioenergy production is presented in Table 8.

In addition to phytoremediation and bioenergy production, the energy crops also help in carbon sequestration and biodiversity management due to their giant structure, longer life cycle, and high above- and below ground biomass, which favors soil microfauna and shelters invertebrates and birds and flora (Blanco-Canqui 2010). They improve soil characteristics by improving soil organic matter, soil aggregation, water retention, hydraulic conductivity, macroporosity, nutrient recycling, and storage and fluxes of water, air, and heat and thus, reduce water and wind erosion (Blanco-Canqui 2010). They also improve water quality by reducing the off-site transport of metal pollutants and thus, reduce the risks of water pollution (Blanco-Canqui 2010). However, the utilization of energy crops in phytoremediation has some distinct disadvantages, which includes: (a) scarcity of agricultural land and threat to food production; (b) diverse agro-climatic conditions for cultivation and difficulty in producing biofuels for the entire year; (c) long maturation phase especially in case of Jatropha that discourage small farmers for its cultivation on agricultural field; (d) air pollution and health hazards due to harmful gaseous emissions during the burning of contaminated biomass; and (e) cost involved in production, growth, transportation, and storage of biomass and its processing. The introduction of a new plant to the agricultural field for phytoremediation and bioenergy production may negatively affect the ecosystem by land use changes, biodiversity and nutrients loss, low yield and finally food security issues (Pandey et al. 2016). Moreover, issues related to the transfer of toxic metals during the burning of contaminated biomass are the major constraints associated with bioenergy production (Gomes 2012). Hence, a thorough life cycle assessment (LCA) is required to fully understand the potential hazards of using contaminated biomass for bioenergy production with low environmental impacts. Further, the quality assessment of produced biofuels and evaluating the suitability of energy crops for biofuel production and phytoremediation of metals-contaminated sites are required before applying at the field scale.

Further, more volume reduction of contaminated biomass is required for its safe disposal and commercial success of phytoremediation. Several methods such as incineration, compaction, ashing, gasification, pyrolysis, direct disposal, and liquid extraction have been suggested for the post-harvest treatment and biomass disposal (Ghosh and Singh 2005; Gomes 2012). Among the available methods, incineration (smelting) is the most acceptable eco-friendly and economically feasible method. It reduces the expenditure for the transportation of biomass but increases the risks of toxic metals leaching to a deeper layer of soil leading to groundwater pollution (Mahar et al. 2016). Pyrolysis can also be a promising method for biomass disposal as pyrolysis product (i.e. biochar) is a source of ore/metal concentrate that could be utilized for the separation and recovery of metals i.e. phytomining and thus, can generate revenue (Ghosh and Singh 2005; Paz-Ferreiro et al. 2014; Dilks et al. 2016). The use of biochar as an additive to soil could also help to sequester carbon and thus, lower the deleterious effects of human-induced climate change due to CO_2 emissions (Paz-Ferreiro et al. 2014). However, no single method is effective to date and researches are underway to find the best disposal method for the metals contaminated biomass. An integrated concept coupling phytoremediation with bioenergy production from contaminated biomass and subsequent metals recovery has been also proposed for the sustainable remediation process (Jiang et al. 2015).

12   Field Experiences

Despite proven success in the laboratory and academics, phytoremediation is still struggling to get a jump from laboratory to field for commercial success. Most of the phytoremediation researches are currently laboratory-based, where conditions are actually different from those in the field. At the field scale, phytoremediation is restricted by many factors such as low metals bioavailability, slow plants growth rate and biomass, reduced metal accumulation and tolerance etc. and several technical difficulties, which need to be catching up (as discussed in section 7). Thus, the selection of suitable plant species for the phytoremediation of HM-contaminated sites is not an easy task. In addition, due to its time-taking nature, remediation industries/companies usually lost their interest in phytoremediation technology to take it up for commercial applications. Applying transgenic plants in phytoremediation is extremely challenging due to

their highly invasive nature and risk of contaminating non-target species with their pollens, but may well remediate the contaminated sites. However, strict US or western countries regulations on their release for field applications, lengthy environmental impact assessment and high cost (approx. $100-150 million) and long time (approx. 10 yrs.) period are the key constraints to get a GEP to market (Beans 2017). Lack of clear-cut understanding about phytoremediation among remediation practitioners is also a key concern. Phytoremediation is being investigated at field scale worldwide. Some updated examples from recent field studies with field experiences are summarized in Table 9.

Reddy et al. (2017) conducted a field trial on the phytoremediation of HMs and polyaromatic hydrocarbons (PAHs)-contaminated slag fill site (Big Marsh, Calumet region, near Chicago, IL, USA) for three years. According to the study, there was no significant decrease in HMs concentrations in soil (no phytoextraction), but HMs were immobilized by native grasses in combination with compost amendment applied to the soil (phytostabilization). However, PAHs were well-degraded (rhizodegradation) and thus, reduced the risk of contaminants to public and environment. Doni et al. (2015) remediated the polluted marine sediments at pilot scale (Port of Livorno, Central Italy) using three selected plants (*Paspalum vaginatum* Sw., *Tamarix gallica* L., and *Spartium junceum* L.) in association with compost to remove HMs (Zn, Cu, Cd, Ni, and Pb). However, Ni and Pb were the lowest translocated metals and the process was largely considered phytostabilization and phytoextraction to a lesser extent. Khaokaew and Landrot (2014) remediated the Cd-contaminated agricultural field (Mae Sot contaminated field, Mae Sot District, Tak Province, Thailand) using Cd-hyperaccumulating plants except for *Chromolaena odorata*, within two months (phytoextraction) under greenhouse condition. Willscher et al. (2013) performed a field study to remediate uranium and other HMs such as Al, Ni, and Zn from a uranium mining site (Gessenwiese, Ronneburg, Eastern Thuringia, Germany) for 14 months using hyperaccumulators, *Helianthus annuus, Triticale* and *Brassica juncea* in association with calcareous topsoil, mycorrhiza, and bacterial culture and harvested plants biomass was utilized for energy production. According to experience, a very low metal accumulation was reported in case of *Triticale; H. annuus* accumulated high Al whereas *B. juncea*

accumulated high Zn and Ni (phytoextraction). Vamerali et al. (2009) reported that phytoextraction of elevated amounts of metals or As from Torviscosa cinder waste site using woody plants, *Populus* and *Salix* sp. is not feasible. This was largely due to low productivity and low levels of translocation of metals from roots and despite an apparent high mobility of Pb and Zn. Thus, the selection of suitable plants is critical for the effective phytoremediation of HMs-contaminated sites.

Further, more long-term field studies are required to document time and cost data for the economic evaluation of *in-situ* phytoremediation at the field scale. Phytoremediation (phytoextraction) is currently under investigation in the US Environmental Protection Agency (U.S. EPA) supported Superfund Innovative Technology Evaluation (SITE) programme (http://www.epa.gov/superfund/sites) to fully understand its economic feasibility, better performance, and wider acceptance. Moreover, phytoremediation is also under investigation in the EU demonstrative projects such as Phytosudoe (http://www.phytosudoe.eu/) or Life RiverPhy

(http://liferiverphy.eu/web/en/) to evaluate its success in environmental decontamination at the field scale.

13   Emerging Phytotechnologies

Phytotechnology is an emerging field that implements solutions to scientific and engineering problems using plants to control and minimize environmental pollution. Phytotechnologies may provide more efficient alternatives to traditional cleanup methods because of their low capital costs and maintenance requirements, high success rates, end-use value, and aesthetic nature. Some phytotechnologies associated with phytoremediation are briefly discussed below:

13.1   *Phytomining*

It is a plant-assisted mining and recovery of precious metals from the ash of combusted-contaminated biomass and thus, can generate revenue. If it applied to the agricultural field, is termed as agro-mining (Sheoran et al. 2009; Mahar et al. 2016; Jiang et al. 2015). Bioenergy generation and less SO_x emission due to the low sulphur content of bio-ores are the additional eco-environmental benefit of phytomining as compared to conventional mining technologies (Ali et al. 2013). It may be limited by the plant's

phytoextraction efficiency and the market price of metals to be processed. It is more suitable for Au, Tl, Co, and Ni due to their high price and concentration in biomass (Mahar et al. 2016). For instance, it has been commercialized for Ni because hyperaccumulator plants such as *Alyssum murale* and *Alyssum corsicum* can accumulate 400 kg Ni ha^{-1} with a production cost of \$250-500 ha^{-1} (Ali et al. 2013). Hence, it is useful for the treatment and management of Ni-contaminated sites. Another successful case study on the phytomining is the use of hyperaccumulator, *Berkheya coddii* for the phytoremediation Nicontaminated soils near an industrial plant in Rustenburg, South Africa (Antony et al. 2015). They reported a high yield of 20t/ha and active Ni absorption 2-3% with metal accumulation in the ash (15%) makes its profitable for the repeated extraction process. The profitability of Ni phytomining using *B. coddii* on Ni-rich serpentine soils (Australia) is estimated at 11,500AU\$/ha/yield and the profitability of Au phytomining using *B. juncea* is about 26,000AU\$/ha/yield (Mahar et al. 2016). However, the high market price of uranium and its low concentration (100 mg/kg) in the biomass (10 t/ha) of *Atriplex* species makes the phytomining unprofitable (Sheoran et al. 2009).

Phytomining can be more economically feasible if combined with bioenergy production and sale of C-credit could be a possible benefit (Mahar et al. 2016). Applying energy crops for the phytoremediation of contaminated sites could also pave the way for economical phytomining of valuable metals. According to a study, the cultivation of energy maize in the Campine region of Netherlands and Belgium could result in the generation of 29000038000KV of renewable energy per hectare (Meers et al. 2010). It may reduce the need for coal-powered energy and minimize the CO_2 emission up to 21 tons/ha/year. However, phytomining is not successful in the northern regions of the world because of low plant productivity under harsh weather conditions. It is mainly suitable for the treatment and restoration of the disturbed soils, mine tailing waste and mining sites in tropical regions and has been accepted by public and commercial enterprises. Thus, phytomining could be an economic incentive for low-cost metal recovery and pollution control.

13.2　Constructed Wetlands

The use of constructed wetlands (CWs) for wastewater treatment and management has been increasingly recognized worldwide. CWs are the man-engineered systems constructed to utilize the natural processes of aquatic macrophytes with their associated microbial assemblages for wastewater treatment within a more controlled environment (Stottmeister et al. 2003; Khan et al. 2009). CWs are mainly vegetated with different wetland plants having high biomass, fast growth rate and metal accumulation capacity such as *Phragmites australis, Typha latifolia, Canna indica, Stenotaphrum secundatum, Scirpus americanus, Scirpus scutus, Iris pseudacorus* etc., for the treatment of metal-rich wastewaters (Bharagava et al. 2017c). CWs have been proved to be successful in the removal of a variety of organic and inorganic pollutants such as metals, nutrients and a wide range of micro-pollutants, such as pharmaceutical and personal care products, and also fecal indicator bacteria and pathogens (Zhang et al. 2015a). The pollutants removal efficiency of CWs mainly depends on wastewater treatment rate, organic loading rate, hydrologic regime, hydraulic retention time, operational mode, and vegetation type (Zhang et al. 2015a). The application of CWs in pollutants removal from wastewaters has been recently reviewed by many workers (Zhang et al. 2014; 2015a; Bharagava et al. 2017c).

　　CWs may provide many ecological and economic benefits such as require low capital investment for construction, low electricity for operation, less maintenance, provides wildlife habitat and human recreational opportunities, and a reuse and recycling option for the wastewater treatment facility. CWs are more favored in developing countries due to easily available and less costly land and tropical environment, which help to flourish the microbial communities responsible for the degradation/detoxification of organic and inorganic contaminants in wastewaters (Zhang et al. 2015). Thus, the increasing use of CWs can successfully treat/detoxify HM-rich wastewaters and solve various water quality issues in the world. Integrating CW plants with a microbial fuel cell (MFC) for wastewater treatment and electricity generation could be an innovative approach for the improved degradation of pollutants. According to a recent study, a maximum current density of 55 mA m^{-2} could be achieved during the removal of hexavalent chromium

(Cr^{6+}) from solution with greater removal efficiency (up to 90%) in an integrated plant-microbial fuel cell (PMFC) system planted with a wetland plant, *Lolium perenne* (Habibul et al. 2016). Moreover, CWs may have great potential for bioenergy production and carbon sequestration, if planted with energy crops. According to a recent study, the incineration of harvested biomass (16,737 kg with C content:

6185 kg) of *Ludwigia* sp. and *Typha* sp. recovered from a subtropical constructed wetland could produce 11,846 kWh energy for one month (Wang et al. 2011). Currently, researches are underway to expand the scope and efficacy of CWs for treatment of metals-contaminated wastewaters. However, future research should be focused on the following points: (a) understanding of microbiological dynamics and correlation of biological and non-biological processes in CWs; (b) knowledge of the dynamics of nutrient cycle to understand the fundamental processes of greenhouse gas emission in CWs; and (c) understanding of microbial community and plant-microbe interactions to know the underlying mechanism of pollutants removal in CWs (Carvalho et al. 2017).

14 Challenges and Future Research Prospects

Phytoremediation has untapped potential to apply in developing countries because of its low-cost and solar-driven nature. However, its field applicability gets limited by low metals phytoavailability, biomass, slow growth rate, and unavailability of target metal hyperaccumulators. Such drawbacks together span a long time-period for phytoremediation to achieve the desirable remediation goal. Therefore, phytoremediation requires a high cost for treatment, safety, and liability of risks involved because long time also adds additional cost while evaluating economic feasibility at the field (Maestri and Marmiroli 2011; Conesa et al. 2012; Mahar et al. 2016). The application of synthetic chelators to achieve higher metal accumulation by plants can also be costly and may lead to undesirable environmental consequences such as disruption of physico-chemical properties of soils by dissolving mineral components; toxicity to soil microorganisms and plants, and unacceptable leaching to groundwater. Hence, the chelate-assisted phytoremediation of HMs-contaminated sites is seems to be impractical. For instance, EDTA costs $30,000 ha^{-1} to accumulate 10 g Pb kg^{-1} dry weight in shoots as well as more readily degradable chemicals are also sometimes very costly

(Chaney et al. 2007; Stephenson and Black 2014). The field applications of metal mobilizing amendments such as EDTA have been also banned in many European countries (Vangronsveld et al. 2009). The growth of plants during phytoremediation leads to changes in soil (e.g., changes in pH, increases in organic acids) that can make metals more bioavailable to the food chain before they can be remediated (Gerhardt et al. 2016). Thus, pose environmental risks that can negative some of the positive effects of phytoremediation. The accumulation of inorganic contaminants *in planta* can lead to their re-release, or release of their toxic forms, into the soil *via* leaf litter. For instance, litter from *Populus alba* that accumulate high levels of Cd and Zn in leaves is deemed to be problematic in the environment (Hu et al. 2013). Moreover, metals that are phytostabilized are not removed from the soil and ever-changing soil conditions may lead to contaminant re-release in the environment and hence, long-term monitoring of the site may be required to avoid land use changes in future (Gerhardt et al. 2016). Phytoremediation efficiency may also be improved by applying genetically modified plants, but such application may also increase the cost of phytoremediation because contaminated sites require greater maintenance, monitoring and disposal of biomass due to strict existing government regulations on their use in the field (Maestri and Marmiroli 2011; Stephenson and Black 2014). The lack of funding from public and private sector agencies for supporting further phytoremediation research is also a major challenge.

Despite several challenges, phytoremediation remains a promising technology with lots of prospects for future research. In recent years, increasing use of phytotechnologies advancing means of phytoremediation by integrating ecological engineering using plants. Phytotechnologies like CWs may provide an eco-technological solution for pollutants removal from wastewaters and income can be generated, if vegetated with local plants such as common reeds, *Phragmites australis,* and elephant grass, *Pennisetum purpureum*, which are being used to produce goods (Stephenson and Black 2014). The development of CWs for wastewater treatment in developing countries is not well reported as in the case of European and American countries. But, Putrajaya wetland in Putrajaya city of Malaysia represents an excellent example of the commercial success of CWs in developing countries (Mahmood et al. 2013; Stephenson and Black 2014).

Fluctuating redox conditions could be helpful in microbial precipitation of iron and sulphate and enhanced degradation of organic pollutants in the rhizosphere of aquatic macrophytes. Thus, it represents an innovative strategy to overcome the limitations of biotechnology and synthetic chelators (Stottmeister et al. 2003; Stephenson and Black 2014).

Exploiting transpiration as a phytotechnology in phytoremediation could be an excellent strategy to control migration of subsurface water along with metal pollutants and is termed as hydraulic control or phytohydraulics (Robinson et al. 2003; Stephenson and Black 2014). Evapotranspiration caps (ETCs) are of great significance in pollution control and management. ETCs are the vegetation cover created over the polluted matrix to prevent the migration of contaminated water from it. These do not destroy or remove contaminants, but prevent the spreading of contaminants and thus, protect people and wildlife (https://www.epa.gov/remedytech/citizens-guideevapotranspiration-covers). ETCs are inexpensive as compared to typical prescriptive covers and can save up to \$120 000-180 000 ha^{-1} area. However, the testing, modeling, and monitoring of these systems may increase the final cost (Stephenson and Black 2014). "Ecolotree cap" of USA represent the first successful example of commercial ETC composed of fast-growing and deep-rooted trees that cover landfills and contaminated soils (Ecolotree 2013). Combining dendroremediation (tree as phytoremediator) with phytostabilization could be an excellent strategy for phytomanagement of contaminated soil. It may also increase the tangible value of land by increasing the provision of wood, feed products and bioenergy production (Robinson et al. 2009; Conesa et al. 2012). Endophytic phytoaugmentation could also be a promising phytotechnology for treating contaminated wastewaters. In this technology, the remediating plants are augmented with potential endophytes for the balanced plant-microbe interactions and enhanced remediation efficiency. However, the slow action, season dependent effectiveness and lack of suitable monitoring methods are the key associated constraints that need to be addressed in the future for a successful application (Redfern and Gunsch 2016).

Combining phytoremediation with electrokinetic remediation (using low-voltage direct electric current to remove organic and inorganic pollutants from contaminated medium) could be an excellent strategy to

enhance the metals mobility in contaminated soil and facilitate their plant uptake and thus, phytoremediation. For instance, Mao et al. (2016) evaluated the feasibility of electrokinetic remediation coupled with phytoremediation to remove Pb, As, and Cs from contaminated paddy soil. Results revealed that the solubility and bioavailability of Cs and As were significantly increased by the electro-kinetic field (EKF) and thereby, lower the pH of contaminated soil. Furthermore, they observed that EKF significantly enhanced the bioaccumulation of As and Cs in plant roots and shoots and thus, enhanced phytoremediation efficiency. The optimization of electrical parameters such as electrical field intensity, current application mode, the distance between the electrodes, and stimulation period and their effect on the mobility and bioavailability of HMs are the associated key challenges (Mao et al. 2016). However, the application of electrokinetic-phytoremediation for the mixed contaminants (organic and inorganic) is also not reported so far (Cameselle et al. 2013). Linking phytomining with conventional mining technologies should also be used for the selective recovery of precious metals from contaminated soils to achieve commercial success (Robinson et al. 2009; Sheoran et al. 2009; Stephenson and Black 2014). Thus, phytotechnologies may provide a way for the sustainable management of HM-contaminated sites.

15 Innovative Ideas and Suggestions for Successful Phytoremediation Practices and Applications at HM-Contaminated Sites

In this section, the innovative ideas and constructive suggestions for the greater acceptance of phytoremediation to be effectively applied for the treatment and management of HM-contaminated sites are provided as below (Gerhardt et al. 2016):

(a) More long-term field studies should be supported by public and private funding agencies and published in the refereed journals indicating that whether or not the remediation was sufficient to meet regulatory compliance at a contaminated site. It will provide some assurance for site managers to choose appropriate remediation options at a given site.

(b) Authors should indicate cost data and estimate of any valorization of biomass in published field studies that allow cost comparison with conventional remediation methods and provide a certainty for site managers to choose appropriate remediation options at a given site.

(c) The low-cost use, wide-applications, and social benefits of phytoremediation should be publicized in social media (*via* YouTube, Facebook, Twitter, and blogs) in addition to scientific journals to educate and engage stakeholders.

(d) Formal (*via* conference presentations) and informal (*via* lunch) meetings with industry persons and stakeholders should be made to discuss its potential and deployment in managing contaminated sites.

(e) The terminology relevant to phytoremediation should be standardized and simplified to make its commercial image/brand, engage non-remediation practitioners, and develop products, services, and technologies.

(f) The cost of landfilling should be increased as it is a less desirable option for the management of contaminated soils. It will inflate the cost and encourage the private sector to explore phytoremediation and other options.

(g) High biomass plants can be used in conjunction with microorganisms (rhizobacteria, ectomycorrhiza, and endophytes) to shorten the time frame and cost required for phytoremediation and allowing rapid turnover of the land for re-use.

(h) Decision support tools (numerous models and decision trees) can be employed to assess the applicability (costeffectiveness and the likelihood of meeting regulatory criteria) of phytoremediation for a given site.

(i) Avoid overselling phytoremediation technology in terms of deployment at contaminated sites because it is not a quick remediation and may not produce satisfactory results at the initial stage. It requires optimization of biology and improvement in soil quality and then can be successfully deployed at the contaminated site.

16 Conclusions

(a) HM pollution in the environment and associated toxicity in living beings is of serious eco-environmental concern.

(b) Phytoremediation is an emerging eco-sustainable and clean-green solution for the eco-restoration of HMs contaminated sites.

(c) Ecological roles of metal hyperaccumulation in plants are still unclear and required to clearly understand the phytoremediation mechanism.

(d) Selecting target plants among known metal hyperaccumulators and exploring new plants for successful phytoremediation is an ongoing challenge.

(e) Investigations on synthetic chelators induced toxicity in remediating plants and fate, dynamics and decomposition of metal-chelators complexes in the rhizosphere are required. The use of cheaper, eco-friendly, non-toxic and degradable organic soil amendments are recommended to minimize environmental pollution, reduce toxicity to remediating plants and enhance phytoremediation efficiency.

(f) Inoculation of plants with associated microbes (such as PGPRs and endophytes) exhibiting multiple traits could be an excellent strategy to enhance metals detoxification in the rhizosphere. A clear-cut understanding of plantmicrobe-metal-soil interactions is crucial for microbe-assisted phytoremediation of HM-contaminated soils.

(g) Genetic engineering of metal accumulating plants and associated microbes with required traits could be a very useful strategy for the enhanced phytoremediation but, associated risks should also be considered before field application.

(h) Linking energy crops with phytoremediation could be an economic incentive for biofuel/energy production and metal recovery with many eco-environmental benefits; however, quality assessment (free from toxic metals) of produced biofuels is strictly advised.

(i) Exploiting stress tolerant medicinal and aromatic plants for metals phytoremediation could be an economically viable approach; however, some are edible plants and thus, their use under strict monitoring is recommended for public health protection.

(j) Search for new and re-evaluating existing methods for the post-treatment of contaminated biomass (processing, volume reduction, and safe disposal) is suggested to gain better economic returns.

(k) Integrating phytoremediation with phytotechnologies or other remediation methods will be helpful in the commercial success of phytoremediation.

(l) Input from different field of science, engineering, and technology is required to support the multidisciplinary research in phytoremediation.

(m) More long-term field trials are required to document time and cost data to provide recommendations and convince regulators, decision-makers, and the general public about the low-cost applicability of phytoremediation to contaminated sites and for better acceptance in remediation industries.

Conclusively, phytoremediation is an eco-technological solution for HM pollution control and management and thus, promotes sustainable development. However, in future, phytomining (i.e. phytoextraction and recovery of precious metals (Au and Ni)) may successfully eliminate or reduce the need of conventional mining at large scale and thus, can generate revenue and wide employment opportunities. Further, performance evaluation, complete utilization of by-products and overall economic feasibility would always be the key criteria for global acceptance of phytoremediation technologies in waste management industries.

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Table Caption:

Table 1 Sources of contamination and toxicity profile of heavy metals

Table 2 Description of phytoremediation mechanisms and applications

Table 3 Hyperaccumulator plants (with metal accumulation capacity) employed for phytoremediation

Table 4 Halophytes and medicinal plants reported in phytoremediation of heavy metal-contaminated soils

Table 5 Recent studies on microbe-assisted phytoremediation of heavy metal-contaminated soils

Table 6 Recent studies on transgenic plants for enhanced phytoremediation of heavy metals

Table 7 Recent studies on genetically engineered bacteria-assisted phytoremediation of heavy metals

Table 8 Energy crops for sustainable phytoremediation of heavy metal-contaminated soils and bioenergy production

Table 9 Recent field studies on phytoremediation of heavy metal-contaminated sites

Figure caption:

Fig. 1 Trophic transfer of toxic HMs from soil to plants to humans and organism's food to humans and their toxicity

Fig. 2 A pictorial representation of different phytoremediation techniques

Fig. 3 Relationships among the factors affecting phytoremediation efficiency

Table 1 Souces of contamination and toxicity profile of heavy metals. Adapted from Sarwar et al. (2016); Dixit et al. (2015); Jaishankar et al. (2014); Ali et al. (2013); Yadav (2010)

Heavy metal	Standard limit		Sources of contamination	Environmental hazrads	Toxic effects	
	Water (mg/l)	Soil (mg/kg)			Plants	Human/animlas
Arsenic (As)	0.01	1 - 50	Pesticides and wood preservatives	Water and soil pollution	Analog of phosphate (P) and hence, compete for the uptake of carriers (P/As) in root plasmalemma and thus, disrupt phosphate-dependent metabolism	Analogue of phosphate and hence, affects oxidative phosphorylation and ATP synthesis, the sensation of "pins and needles" in hands and feet
Cadmium (Cd)	0.003	0.01 - 0.7	Paints and pigments, plastic stabilizers, electroplating, incineration of cadmium containing plastics, phosphate fertilizers	Water and soil pollution	Chlorosis, browning of root tips, reduced seed germination, growth photosynthesis, water, nitrate, and nutrient uptake, and ATPase activity, Fe(II) deficiency and ultimately leads to death	Carcinogenic, mutagenic, teratogenic; endocrine disruptor, hypercalciuria, and *Itai-Itai* disease
Lead (Pb)	0.01	2 - 200	Aerial emission from combustion of leaded petrol, battery manufacture, herbicides and insecticides	Water and soil pollution	Chlorosis, reduced seed germination, growth, biomass, photosynthesis, nutrients and water uptake, and transport, alter membrane permeability, induce abnormal morphology, oxidative stress (ROS generation) in plants and inhibit enzymatic activity at the cellular level by reacting with their sulfhydryl groups	Impaired development, reduced intelligence, short-term memory loss, insomnia, anorexia, encephalopathy, disabilities in learning and coordination problems, the risk of cardiovascular disease, foot drop/wrist drop (palsy) and nephropathy
Chromium (Cr)	0.05	1 - 1,000	Tanneries, steel industries, fly ash, dyes, and pigments	Water and soil pollution	Chlorosis, membrane damage, nutrient imbalance, wilting of tops, and root injury, reduced seed germination, growth and development, photosynthesis, and enzymatic activity	Highly toxic proven carcinogen as identified by IARC, WHO, ATSDR, and USEPA; hair loss, pulmonary fibrosis (lung scarring), lung cancer and damage to the kidney, circulatory and nerve tissues
Mercury (Hg)	0.001	0.01 - 0.3	Release from Au-Ag mining and coal combustion, surgical instruments, medical waste	Water and soil pollution	Interfere with mitochondrial activity and induces oxidative stress by triggering ROS generation and thus, disrupt membrane lipids and cellular metabolism	Possible human carcinogen (methyl-Hg) as established by USEPA, anxiety, Minamata, autoimmune diseases, depression, difficulty with balance, drowsiness, fatigue, hair loss, insomnia, irritability, memory loss, recurrent infections, restlessness, vision disturbances, tremors, temper outbursts, ulcers and damage to brain, kidney and lungs, neurasthenia (neurotic disorder) and parageusia (metallic taste)
Copper (Cu)	2.00	2 - 100	Pesticides and fertilizers	Water and soil pollution	Chlorosis, reduced growth, induces stress (ROS generation) and thus, disturb metabolic pathways and damage to macromolecules, exert cytotoxic effects, and ultimately injury to plants	Wilson's disease, brain and kidney damage, liver cirrhosis, chronic anemia, stomach and intestine irritation, and even death
Nickel (Ni)	0.02	5 - 500	Industrial effluents, kitchen appliances, surgical instruments, steel alloys, automobile batteries	Water and soil pollution	Reduced seed germination, chlorosis, necrosis, nutrient imbalance, ion imbalance particularly K+, alteration in cell membrane functions, lipid composition and H-ATPase activity of the plasma membrane	Hematotoxic, immunotoxic, neurotoxic, genotoxic, reproductive toxic, pulmonary toxic, nephrotoxic, and hepatotoxic, allergic dermatitis (itching), cancer of the lungs, nose, sinuses, throat, and stomach, hair loss and defects in infants, cardiovascular and musculoskeletal system

Standard limit of heavy metals in drinking water according to WHO drinking water standard (1993) for standard setting and drinking water safety. Access online at:
https://www.lenntech.com/applications/drinking/standards/who-s-drinking-water-standards.htm
§Common range of heavy metals in soil according to Lindsay (1979). Access online at: http://www.occeweb.com/og/metals-limits.pdf

*Abbreviations: IARC: International Agency for Research on Cancer; WHO: World Health Organisation; ATSDR: Agency for Toxic Substances and Disease Registry; USEPA: United States Environmental Protection Agency; ROS: Reactive Oxygen Species; LDL: Low-Density Lipoprotein; HDL: High-Density Lipoprotein

Table 2 Description of phytoremediation mechanisms and applications

Phytoremediation processes	Definition	Mechanism	Pollutants	Applicability	Benefits	Comments/Issues
Phytoextraction or phytoaccumulation or phytoabsorption or phytosequestration	Plants remove metal pollutants from contaminated sites via plants root absorption, sequester/concentrate in above-ground harvestable plant parts	Hyper-accumulation	Pb, Cd, Zn, Ni, Cu, Pb, radionuclides, pentachlorophenol, aliphatic compounds (short chained)	Contaminated soil/sites, water, wastewaters	Abundant biomass in short time, reduced soil erosion and cost-effective, wide application prospects	Slow process, contaminant concentration is important and its depend on the depth of contamination, the risk of metals leaching and thus, ground water pollution, require post-harvest treatment for contaminated biomass volume reduction, metals recovery (i.e. phytomining) and bioenergy production
Phytofiltration or rhizofiltration	Plants concentrate and precipitate metal pollutants in low concentration from the aquatic environment in their roots	Rhizosphere accumulation	Pb, Cd, Zn, Ni, Cu, Radionuclides (Cs, Sr, U), hydrophobic organics, and radionuclides	Contaminated water and wastewaters	Cleanup of polluted surface water, industrial wastewaters, and agricultural runoff	Plants roots act as filters for cleanup of polluted water/wastewaters; less generation of secondary waste and minimize the need of further disposal, if terrestrial plants are used due to high biomass as compared to aquatic plants (highly species specific), long-term maintenance depends on the type of contaminant and contamination depth
Phytostabilization or phytoimmobilization or phytotransformation	Plants immobilize or inactivate metal pollutants at their place involving absorption by roots, adsorption onto roots and precipitation, complexation and metal valence reduction in rhizosphere e.g. reduction of Cr^{6+} to Cr^{3+}	Precipitation, complexation and metal valence reduction	Pb, Cd, Zn, As, Cu, Cr, Se, U, polycyclic aromatic hydrocarbons (PAHs), polychlorinated biphenyl (PCBs), dioxins, furans, pentachlorophenol, DDT, Dieldrin	Contaminated soil/sediments and sludge	Ecologically efficient, stabilization of contaminated medium without disposal of contaminated biomass, reduces soil erosion, applicable in the field and mine polluted areas	Not a permanent solution because plants only limit mobility/bioavailability of metal pollutants at the place and hence, the site cannot be used for plant growth
Phytovolatilization or phytoevaporation	Plants uptake metal pollutants through roots in low concentration, modify/transform them into less toxic form and subsequently transpire/volatilize into the atmosphere through stomata	Volatilization or evaporation by leaves	Chlorinated solvents like carbon tetrachloride, trichloroethylene, methylene chloride, tetrachloroethylene, carbon tetrachloride, 1,1,1-Trichloroethane, Hg (mercuric ion), Se	Contaminated wastewaters, soil, sediments, and sludges	Environmental cleanup without harvesting plants and biomass disposal	Suitable for Hg^{2+} removal; limited in case of Se, elemental Hg and As due to their gaseous forms; most controversial because there may be a chance of staying toxic metals in air and thus, no control over migration (air pollution); Re-deposition of pollutant back into the ecosystem by precipitation (elemental Hg)
Phytodegradation	Plants breakdown/convert highly toxic organic pollutants into less toxic forms through the action of enzymes secreted within plant tissues and released in the rhizosphere	Degradation in plant tissues	DDT, PAHs, bisphenol A, organophosphorus compounds	Contaminated soil, sediments, sludges, groundwater, surface water, and wastewaters	Biodegradation of various recalcitrant pollutants in the rhizosphere	Key enzymes for degradation are nitroreductase, dehalogenase, oxygenase, peroxidase, nitrilase, nitroreductase, and laccase, depends on factors such as concentration and composition, plant species, and soil conditions
Rhizodegradation or rhizoremediation or phytostimulation	Plants breakdown/convert highly toxic organic pollutants into less toxic forms though enzymatic activity of rhizospheric microorganisms	Degradation in rhizosphere	Atrazine, ammunition wastes, petroleum hydrocarbon, PCBs, PAHs, TCE, diesel fuel	Contaminated soil, sediments, sludges, groundwater and wastewaters	Release of organic acid, rhizosphere enhances biodegradation, metabolic products are further utilized by microorganisms in rhizosphere	Depends on the action of root exudates and enzymes; increased activity of rhizosphere microbes

Table 3 Hyperaccumulator plants (with metal accumulation capacity) employed for phytoremediation

Plant species	Common name	Family	Polluted medium	Metal	Metal accumulation capacity (mg kg⁻¹ DW)	Phytoremediation mechanism and metal accumulation compartment	Reference
Noccaea caerulescens	Blue stool	Brassicaceae	Water	Pb	1700-2300	Rhizofiltration (aerial parts/root)	Dinh et al. (2018)
Cannabis sativa L.	Hemp	Cannabaceae	Soil	Cd	151	Phytoextraction (above ground plant parts)	Ahmed et al. (2016)
Alyssum markgrafii	Madwort	Brassicaceae	Soil	Ni	4038	Phytoextraction (above ground plant parts)	Salihaj et al. (2016)
Cannabis sativa L.	Hemp	Cannabaceae	Soil	Cu	1530	Phytoextraction (above ground plant parts)	Ahmed et al. (2016)
Tagetes minuta	Southern cone marigold	Asteraceae	Water	As	380.5	Phytoextraction (shoots)	Salazar and Pignata (2014)
Sorghum halepense L.	Johnson grass	Poaceae	Soil	Pb	1406.8	Phytostabilization (reduction in rhizosphere)	Salazar and Pignata (2014)
Betula occidentalis	Water/Red birch	Betulaceae	Soil	Pb	1000	Phytoextraction (shoots)	Koptsik (2014)
Thlaspi caerulescens	Alpine pennygrass	Brassicaceae	Soil	Cd	5000	Phytoextraction (shoots)	Koptsik (2014)
Helianthus annuus	Sun flower	Asteraceae	Soil	Pb	5600	Phytoextraction (shoots)	Koptsik (2014)
Thlaspi caerulescens	Alpine pennygrass	Brassicaceae	Soil	Ni	16,200	Phytoextraction (shoots)	Koptsik (2014)
Brassica nigra	Black mustard	Brassicaceae	Soil	Pb	9400	Phytoextraction (shoots)	Koptsik (2014)
Medicago sativa	Alfalfa/Lucerne	Fabaceae	Soil	Pb	43,300	Phytoextraction (shoots)	Koptsik (2014)
Pteris vittata	Chinese ladder rake	Pteridaceae	Soil	Hg	91.975	Phytoextraction (shoots)	Jianxu et al. (2012)
Corrigiola telephiifolia	Corrigiola (Spanish)	Caryophyllaceae	Soil	As	2110	Phytoextraction (above ground plant parts)	Garcia-Salgado et al. (2012)
Achillea millefolium	Common yarrow	Asteraceae	Soil	Hg	18.275	Phytoextraction (shoots)	Jianxu et al. (2012)
Pteris vittata	Chinese ladder rake	Pteridaceae	Water	As	20,707	Phytoextraction (shoots)	Kalve et al. (2011)
Eleocharis acicularis	Needle spikerush	Cyperaceae	Water	Zn	11,200	Phytoextraction (shoots)	Sakakibara et al. (2011)
Solanum photeinocarpum	American black nightshade	Solanaceae	Soil	Cd	158	Phytoextraction (shoots)	Zhang et al. (2011)
Eleocharis acicularis	Needle spikerush	Cyperaceae	Water	Cu	20,200	Phytoextraction (shoots)	Sakakibara et al. (2011)
Pteris vittata	Chinese ladder rake	Pteridaceae	Water	Cr	35,303	Phytoextraction (shoots)	Kalve et al. (2011)
Eleocharis acicularis	Needle spikerush	Cyperaceae	Water	Cd	239	Phytoextraction (shoots)	Sakakibara et al. (2011)
Pteris vittata	Chinese ladder rake	Pteridaceae	Water	As	20,707	Phytoextraction (shoots)	Sakakibara et al. (2011)
Eleocharis acicularis	Needle spikerush	Cyperaceae	Water	As	1470	Phytoextraction (shoots)	Sakakibara et al. (2011)
Alyssum murale	Yellow tuft	Brassicaceae	Soil	Ni	4730-20100	Phytoextraction (shoots)	Bani et al. (2010)
Alyssum markgrafii	Madwort	Brassicaceae	Soil	Ni	19100	Phytoextraction (shoots)	Bani et al. (2010)
Thlaspi caerulescens	Alpine pennygrass	Brassicaceae	Soil	Cd	3000	Phytoextraction (shoots)	Sheoran et al. (2009)
Haumaniastrum katangense	Copper flower	Lamiaceae	Soil	Cu	8356	Phytoextraction (shoots)	Sheoran et al. (2009)
Rorippa globosa	Globe yellowcress	Brassicaceae	Soil	Cd	>100	Phytoextraction (shoots)	Wei et al. (2008)
Azolla pinnata	Water velvet	Salviniaceae	Water	Cd	740	Rhizofiltration (bioaccumulation)	Rai (2008)
Marrubium vulgare	Common horehound	Lamiaceae	Soil	Hg	13.8	Phytoextraction (shoots)	Luis et al. (2007)

Table 4 Halophytes and medicinal plants reported in phytoremediation of heavy metal-contaminated soils

Halophytes	Metal	Remarks	Reference
Spartina alterniflora	Cu	Accumulation of metal in roots (phytostabilization)	Chai et al. (2014)
Phragmites australis	Cd	Accumulation of metal in roots (phytostabilization)	Nunes da Silva et al. (2014)
Moringa oleifera	Cd	Through leaf extraction (phytoextraction)	Howladar (2014)
Zygosaccharomyces rouxii	Cd, Zn, Cu, & Pb	Extraction and exclusion of heavy metals (phytoextraction)	Li et al. (2013)
Salicornia bigelovii	Se	Se removal up to 251.6 ± 140.5 µg/m^{2}d (Phytovolatilization)	Huang et al. (2013)
Halimione portulacoides	Zn	Accumulation of metal in tissues (phytostabilization)	Andrades-Moreno et al. (2013)
Suaeda salsa	Pb & Zn	Accumulation of metals in roots (phytostabilization)	Wu et al. (2013)
Salicornia ramosissima	Cd	Accumulation of metal in roots (phytostabilization)	Pedro et al. (2013)
Arthrocnemum macrostachyum	Cd	Accumulation of metal in roots (phytostabilization)	Redondo-Gómez et al. (2010)
Commelina communis	Cu	Accumulation of metal in roots (phytostabilization)	Wang and Zhong (2011)
Salicornia brachiata	Cd, Ni, & As	Accumulation metals in roots (phytostabilization)	Xu et al. (2010)
Sarcocornia perennis	Cd, Zn, Cu, & Co	Accumulation of metal in roots (phytostabilization)	Lefèvre et al. (2010)
Medicinal plants	**Metal**	**Remarks**	**Reference**
Lavandula vera L.	Cd, Pb and Zn	Plant is a good phytoremediator for Pb, Cd, and Zn (phytoextraction)	Angelova et al. (2015)
Portulaca oleracea L.	Cr	Significant Cr accumulation in harvestable parts (phytoextraction)	Kale et al. (2015)
Portulaca oleracea L.	Pb, Ni and Zn	Plant species is not suggested for HMs phytoremediation purpose due to inhabitation of plant roots at high metals concentration	Amer et al. (2013)
Ocimum tenuiflorum L., *Ocimum gratissimum* L. and *Ocimum basilicum* L.	As	*Ocimum* spp. can be used as a phytoremediator and at the same time as a source of essential oils under proper regulation	Siddiqui et al. (2013)
Ocimum gratissimum L.	Cd and Zn	Plant species could not be considered as hyperaccumulator due to poor metals translocation capacity from roots to shoots	Chaiyarat et al. (2011)
Artemisia annua L.	As	*A. annua* could be considered as the top potential candidate for cultivation in As contaminated soil	Rai et al. (2011)
Mentha arvensis L., *Mentha piperita* L. and *Mentha citrata* L.	Cr and Pb	The studied species are not hyperaccumulator for Cr and Pb	Prasad et al. (2010)
Nasturtium officinale R. Br	Ni	*N. officinale* can be utilized as a phytoremediator in moderately polluted wetlands (phytoextraction)	Duman and Ozturk (2010)

Table 5 Recent studies on microbe-assisted phytoremediation of heavy metal-contaminated soils

Bacterial strain(s)	Host plant	Heavy metal	Metal conc. in control plant	Metal conc. in inoculated plant	Medium	Beneficial effects	Reference
Enterobacter sp. LC1, LC4, & LC6, *Kocuria* sp. LC2 & LC5, and *Kosakonia* sp. LC7	*Solanum nigrum*	As	152.3 mg kg^{-1}	2000 mg kg^{-1}	Soil	IAA and P-solubilization	Mukherjee et al. (2018)
Pseudomonas libanensis and *Pseudomonas reactans*	*Brassica oxyrrhina*	Cu, Zn	Cu: 93 mg kg^{-1}, Zn: 835 mg kg^{-1}	Cu: 219 mg kg^{-1}, Zn: 1254 mg kg^{-1}	Soil	IAA, ACC deaminase, siderophores	Ma et al. (2016a)
Rhizobium leguminosarum	*Brassica juncea*	Zn	Above ground: 100 mg kg^{-1}, Below ground: 200 mg kg^{-1}	Above ground: 1200 mg kg^{-1}, Below ground: 400 mg kg^{-1}	Soil	Metal chelation	Adediran et al. (2015)
Bacillus pumilus E2S2 and *Bacillus* sp. E1S2	*Sedum plumbizincicola*	Cd	125 mg kg^{-1}	180 mg kg^{-1} (E2S2), 165 mg kg^{-1} (E1S2)	Soil	IAA, ACC deaminase, siderophores, P-solubilization	Ma et al. (2015)
Pseudomonas sp. LK9	*Solanum nigrum*	Cd	230 mg kg^{-1}	292 mg kg^{-1}	Soil	Biosurfactants, siderophores, organic acids	Chen et al. (2014)
Enterobacter sp. JYX7 and *Klebsiella* sp. JYX10	*Polygonum pubescens*	Cd	Root: 65 mg Cd kg^{-1}, Shoot: 38 mg Cd kg^{-1}	Root: 170 mg Cd kg^{-1} (JYX7), 155 mg Cd kg^{-1} (JYX10); Shoot: 50 mg Cd kg^{-1} (JYX7), 56 mg Cd kg^{-1} (JYX10)	Soil	IAA, siderophores, ACC deaminase, P-solubilizaiton	Jing et al. (2014)
Bacillus thuringiensis GDB-1	*Alnus firma*	As	Root:1911 mg kg^{-1}, Shoot:762, mg kg^{-1}	Root: 2162 mg kg^{-1}, Shoot: 845 mg kg^{-1}	Mine tailing waste	ACC deaminase, IAA, siderophores, P-solubilization	Babu et al. (2013)
Rahnella sp. JN6	*Brassica napus*	Pb	Shoot: 22 mg kg^{-1}, Root: 77 mg kg^{-1}	Shoot: 32 mg kg^{-1}, Root: 113 mg kg^{-1}	Soil	IAA, ACC deaminase, siderophores, P-solubilization	He et al. (2013)
Staphylococcus arlettae NBRIEA G-6	*B. juncea*	As	Root: 11 mg kg^{-1}, Shoot: 2 mg kg^{-1}	Root: 12 mg kg^{-1}, Shoot: 70 mg kg^{-1}	Soil	IAA, siderophores, ACC deaminase	Srivastava et al. (2013)
Rahnella sp.	*Amaranthus hypochondriacu, A. mangostamus and S. nigrum*	Cd	55 µg pot^{-1}	130 µg pot^{-1}, 117 µg pot^{-1}, 110 µg pot^{-1}	Soil	IAA, siderophores, ACC deaminase, P-solubilizaiton	Yuan et al. (2013)
Paenibacillus macerans NBRFT5, *Bacillus endophyticus* NBRFT4, and *Bacillus pumilus* NBRFT9	*Brassica juncea*	Ni	300 µg plant^{-1}	Combined treatment: 950 µg plant^{-1}	Mix. of fly ash and press mud	Siderophores, organic acids, protons and other non specified enzymes	Tiwari et al. (2012)
Pantoea agglomerans Jp3-3, and *Pseudomonas thivervalensis* Y1-3-9	*Brassica napus*	Cu	Shoot: 8 µg pot^{-1}, Root: 7 µg pot^{-1}	Shoot: 25 µg pot^{-1} (Jp3-3), Root: 20 µg pot^{-1} (Jp3-3); Shoot: 25 µg pot^{-1} (Y1-3-9), Root: 15 µg pot^{-1} (Y1-3-9)	Quartz sand	IAA, siderophores, ACC deaminase, P-solubilization	Zhang et al. (2011b)
Azotobacter chroococum and *Rhizobium leguminosarum*	*Zea mays L.*	Pb	290 µg g^{-1}	1329 µg g^{-}	Soil	IAA production increased and soil pH decreased	Hadi and Bano (2010)
Bacilus subtilis, B. cereus, Flavobacterium sp. and *Pseudomonas* sp.	*Orycoprhagmus violaceus*	Zn	Root: 187 mg kg^{-1}, Shoot: 253 mg kg^{-1}	Root: 238-377 mg kg^{-1}, Shot: 290-486 mg kg^{-1}	Soil	ACC deaminase, IAA, siderophores	He et al. (2010)

Table 6 Recent studies on transgenic plants for enhanced phytoremediation of heavy metals

Transgene	Origin	Target plant	Effect(s)/enhanced trait	Reference
OsMT-3a	*Oryza sativa*	*Oryza sativa*	Enhanced tolerance to Cd	Mekawy et al. (2018)
VsCCoAOMT	*Vicia sativa*	*Arabidopsis thaliana*	Enhanced tolerance and accumulation of Cd	Xia et al. (2018)
AtACR2	*Arabidopsis thaliana* L.	*Nicotiana tabacum,* var Sumsun	Plants well survived on culture medium containing 200 μM arsenate, but control plants were barely survived; upon exposure to 100 μM arsenate for 35 days treatment, As content in shoots was significantly lower (28 μg/g d. wt.) than control (40 μg/g d. wt.) whereas in root, it was significantly higher (2400 μg/g d. wt.) than control (2100 μg/g d. wt.)	Nahar et al., (2017)
SaHMA3	*Sedum alfredii*	*Nicotiana tabacum*	Cd tolerance and accumulation	Zhang et al. (2016)
StGCS-GS	*Streptococcus thermophilus*	*Beta vulgaris* L.	Enhanced tolerance and accumulation of different concentrations of Cd, Zn and Cu in shoot with increased biomass, root length and relative growth compared with wild-type plants	Liu et al. (2015)
SaMT2	*Sedum alfredii*	*Saccharomyces cerevisiae/Nicotiana tabacum*	Enhanced tolerance and accumulation of Cd	Zhang et al. (2014)
MerE	E.coli XL1-Blue	*Arabidopsis thaliana* L.	Enhanced tolerance and accumulation of methymercury and mercuric ions in plants as compared to control	Sone et al. (2013)
CYP2E1 and GST	*Homo sapiens*	Alfalfa *(Medicago sativa L.)*	Accumulations of Hg and TCE were increased (3.0-4.2 fold for Hg, 2.1-4.0 fold for TCE) from liquid MS medium with increased resistance upto 50/500 μM of Hg/TCE (trichloroethylene) complex than control	Zhan et al. (2013)
ScYCF1	*Saccharomyces cerevisiae*	*Populus alba* X P. *tremula var. glandulosa,* BH1 *poplar clone*	Enhanced growth, reduced toxicity symptoms, and increased Cd content in aerial tissues from hydroponic solution than control; increased Cd, Zn and Pb content in root than control from mine tailing soil	Shim et al. (2013)
EhMT1	*Elsholtzia haichowensis*	*Nicotiana tabacum*	Enhanced tolerance and accumulation of Cd	Xia et al. (2012)
YCF1	*Saccharomyces cerevisiae*	*Brassica juncea* L.	Seedlings showed 1.3 to 1.6 fold tolerance to Cd stress and 1.2 to 1.4 fold tolerance to Pb stress as compared to wild type (WT) plants (per gram fresh weight) with increased accumulation of metals (1.5 to 2 fold higher Cd(II) and Pb(II) levels than those of WT	Bhuiyan et al. (2011)
tcu1	*Neurospora crassa*	*Nicotiana tabacum* L.	Higher acquisition of copper (up to 3.1 times) in transgenic plants grown in hydroponic medium as compared with control plants with more Cu accumulation when grown in soil, however, supplementation of other divalent cations such as Cd^{2+} and Zn^{2+} did not alter uptake of Cu by transgenic plants	Singh et al. (2011)
tzn1	*Neurospora crassa*	*Nicotiana tabacum* L.	Enhanced accumulation of Zn (up to 11 fold higher) was reported as compared to control, however, no significant difference in uptake of Cd^{2+}, Fe^{2+}, Ni^{2+}, Cu^{2+}, Mn^{2+} and Pb^{2+} between the transgenic and control plants was observed	Dixit et al. (2010)
PsMTA1	*Pisum sativum* L.	*Populus alba* L.	Plant survived at 0.1 mM $CuCl_2$ concentration at which the growth of control was strongly affected; root growth was slightly affected due to ectopic expression of the transgene; ROS accumulated at lower extent in leaf tissues than control with greater tolerance to photooxidative stress and low DNA damage upon exposure to Cu	Balestrazzi et al. (2009)
merP	*Bacillus megaterium*	*A. thaliana* L.	Plant were capable of germination and growth on media amended with 12.5 μM Hg^{2+} accumulating 5.35 μg Hg^{2+}/g of fresh seedling weight	Hsieh et al. (2009)
GSH1	*S. cerevisiae*	*A. thaliana* L.	Cd accumulation increased 4 times higher from media amended with 30 ppm Cd^{2+} and As accumulation increased 2.5 and 4.4 times higher from media amended with 28 ppm AsO_4^{3-} and AsO^{2-}, respectively however, no effect on Cd^{2+}, AsO_4^{3-} and AsO^{2-} tolerance were observed	Guo et al. (2008)
GSH1 and AsPCS1	*S. cerevisiae* and *A. sativum*	*A. thaliana* L.	Roots length increased 2 times longer on media with 50 μM Cd^{2+}, 150 μM AsO_4^{3-} or 50 μM AsO^{2-}; Cd accumulation increased (10 times higher from media with 30 ppm Cd^{2+}) and As accumulation increased (3 and 10 times higher from media amended with 28 ppm AsO_4^{3-} and AsO^{2-}, respectively	Guo et al. (2008)
AtPCS1	*A. thaliana* L.	*B. juncea* L.	Root length increased 1.9 and 1.4 times longer, respectively on media amended with 100 μM Cd^{2+} and 500 μM AsO_4^{3-}, respectively	Gasic and Korban (2007)

Foot note:

[a]Encoded gene products: OsMT-3a: Metallothionein; VsCCoAOMT: V. sativa homologue of Caffeoyl-CoA Omethyltransferase; *GST*: Glutathione S-transferase; *CYP2E1*: Cytochrome P450 2E1; *ScYCF1*: *Saccharomyces cerevisiae* cadmium factor1; SaHMA3: P 1B-type ATPase; *PsMTA1*: Type 2 MT able to bind Cu in planta with strong affinities for other metal ions such as Zn and Cd; *YCF1*: yeast cadmium factor 1; AtACR2: *Arabidopsis thaliana* arsenic reductase 2; SaMT2: Metallothionein; StGCS-GS: *Streptococcus thermophilus* γ-glutamylcysteine synthetase-glutathione synthetase; EhMT1: Metallothionein; *merE*: a broad-spectrum mercury transporter; *MerP*: Hg^{2+}-binding protein encoded within TnMERI1; *GSH1*: Yeast glutathione synthetase; *AsPCS1*: Phytochelatin synthase of garlic; *AtPCS1*: Phytochelatin synthase (PCS) of Arabidopsis; *tzn1*: Zn transporter; *tcu1*: a high affinity Cu transporter

Table 7 Recent studies on genetically engineered bacteria-assisted phytoremediation of heavy metals

Genetically modified bacteria	Modified gene expression	Host plant	Beneficial effects	Heavy metal(s)	Reference
Pseudomonas aeruginosa (Pse-w-MT)	Mouse metallothioneins (MTs)	*Pisum sativum* L.	IAA, siderophore production, and P-solubilization	Cd	Huang et al. (2016)
Pseudomonas putida KT2440	Phytochelatin synthase (PCS)	*Triticum aestivum*	Production of phytochelatins (PCs)	Cd, Hg and Ag	Yong et al. (2014)
Burkholderia sp. HU001 and *Pseudomonas* sp. HU002	pTOM plasmid of *Burkholderia vietnamiensis* BU61	*Salix schwerinii × S. viminalis* cv. Tora	Phytoremediation of mixed Cd-toluene contaminations	Cd-toluene	Weyens et al. (2013)
Meshorhizobium huakuii subsp. rengei strain B3	Iron regulated transporter 1 gene from *Arabidopsis thaliana* (*ATIRT1*)	*Astragalus sinicus*	Production of MTs and PCs, enhance nodule formation	Cu, Cd, Zn and As	Ike et al. (2008)
Meshorhizobium huakuii subsp. rengei strain B3	MTL4 and *AT*PCS		Production of MTs and PCs	Cd	Ike et al. (2007)
Pseudomonas putida 06909	Expression of metal binding peptide (EC20)	*Helianthus Annuus*	Production of metal binding peptide	Cd	Wu et al. (2006)
Meshorhizobium huakuii subsp. rengei strain B3	PCS*AT*		Production of phytochelatins (PCs)	Cd	Sriprang et al. (2003)
Meshorhizobium huakuii subsp. rengei strain B3	Tetrameric human metallothionein (MTL4)	*Astragalus sinicus*	Production of metallothioneins (MTs)	Cd	Sriprang et al. (2002)
Enterobactor cloacae CAL2	EC 4.1.99.4	*Brassica napus*	IAA, ACC deaminase, siderophores and antibiotics	As	Nie et al. (2002)

Table 8 Energy crops for sustainable phytoremediation of heavy metal-contaminated soils and bioenergy production

Plant species	Common name	Family	Target pollutants	Bioenergy/fuel products	References
Arundo donax L.	Giant reed	Poaceae	Phytoremediation of Zn, Cr and Pb contaminated soil	Bioethanol production from biomass	Zucaro et al. (2016); Barbosa et al. (2015)
Phragmites australis L.	Common reed	Poaceae	Phytoremediation of Co, Ni, Mo, Cd, Pb, Cr, Cu, Fe, Mn, Zn, and Hg contaminated sites	Cellulosic butanol production	Cicero-Fernández et al. (2015), Gao et al. (2014)
Cannabis sativa L.	Hemp	Cannabaceae	Phytoremediation of Cu, Cd and Ni contaminated soil	Biogas and solid biofuel production from biomass	Ahmad et al. (2016); Prade et al. (2012)
Eucalyptus sp.	Eucalyptus	Myrtaceae	Phytoremediation of Cu, Zn, Cd and Pb contaminated soil	Carbon sequestration and bioenergy production from biomass	Nirola et al. (2015); Pérez-Cruzado et al. (2011)
Ricinus communis L.	Castor bean	Euphorbiaceae	Phytoremediation of Cd, Pb and DDT contaminated soil	Biodiesel production from seed oil	Zhang et al. (2015b); Huang et al. (2011); Lavanya et al. (2012);
Miscanthus sp.	Silver grass	Poaceae	Phytoremediation of As, Cu, Pb, Ni, Cd, and Zn contaminated water and soil	Bioethanol production from biomass	Bang et al. (2015); Kim et al. (2012)
Jatropha curcas L.	Physic nut	Euphorbiaceae	Phytoremediation of Cd, Pb and Zn contaminated soil	Biodiesel production from seed oil	Chang et al. (2014); Berchmans and Hirata (2008)
Panicum virgatum L.	Switch grass	Poaceae	Phytoremediation of Zn, Cr and Cd contaminated soil	Cellulosic butanol production	Gao et al. (2014); Chen et al. (2012)
Populus sp.	Poplar	Salicaceae	Phytoremediation of Cd, Cu, Fe, Pb and Zn contaminated soil	Thermal and electrical energy from biomass	Baldantoni et al. (2014); Das and Chaturvedi (2009)
Salix sp.	Willow	Salicaceae	Phytoremediation of Cd, Cu, Hg, Pb and Zn contaminated soil	Bioethanol production and electricity generation from biomass	Mleczek et al. (2010)
Zea mays L.	Corn	Poaceae	Phytoremediation of Cd, Pb and Zn contaminated soil	Thermal and electrical energy from biomass	Meers et al. (2010)

Table 9 Recent field studies on phytoremediation of heavy metal-contaminated sites

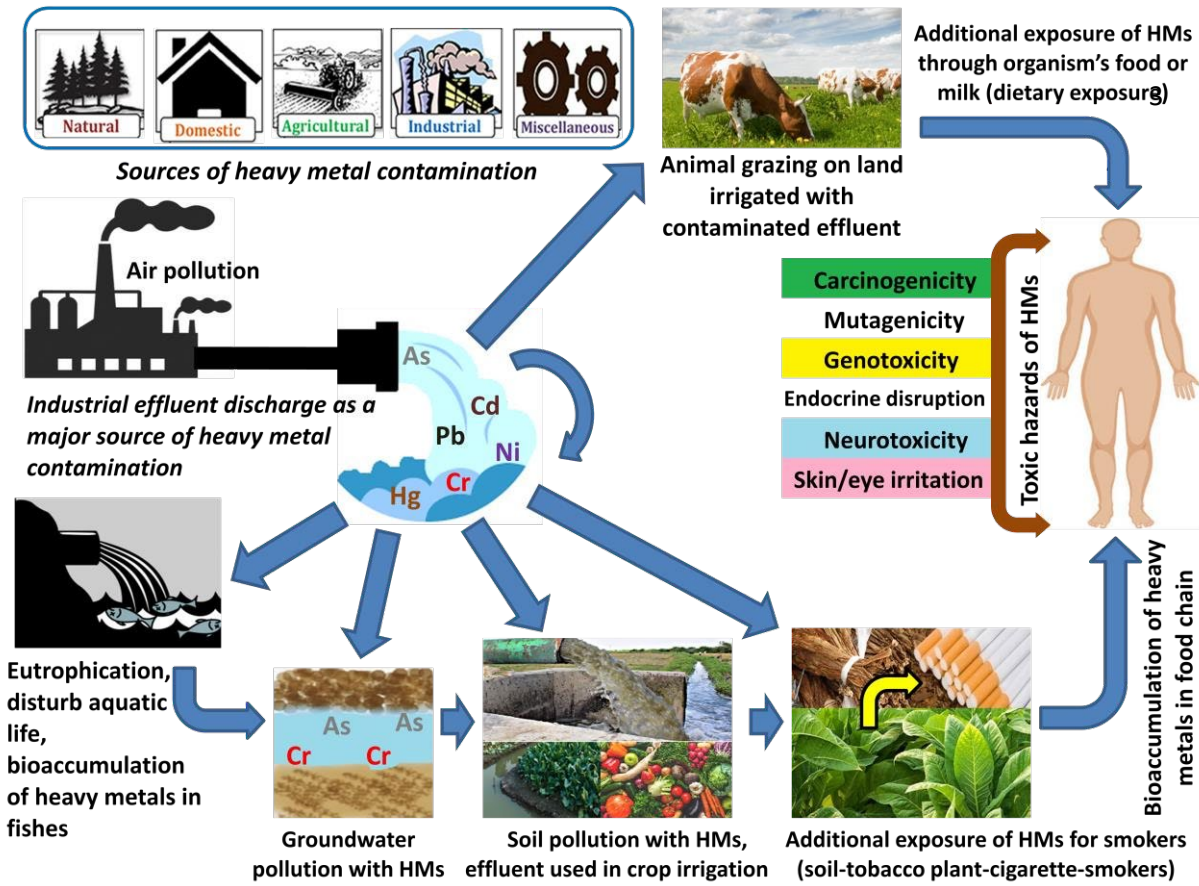| Contaminated field/site	Type of field/site	Metal pollutants	Amendment applied	Plant species	Treatment time	Remediation technique	Field experiences	References
"Big Marsh", Calumet region, near Chicago, IL, USA	Slag fill site	PAHs and heavy metals	Compost	Many plant species (please see paper)	3 years	Rhizodegradation and phytostabilization	Insignificant uptake of HMs by plants, degradation or immobilization of contaminants	Reddy et al. (2017)
Port of Livorno, Central Italy	Polluted marine sediments	Cd, Zn, Cu, Pb and Ni	Compost	*Paspalum vaginatum* Sw.; *P. vaginatum* Sw. + *Spartium junceum* L.; and *P. vaginatum* Sw. + *Tamarix gallica* L.		Phytoextraction and phytostabilization	Best uptake of Cd, Zn, and Cu by plants whereas Ni and Pb were the lowest translocated metals	Doni et al. (2015)
Bouwer, Province of Córdoba, Argentina	Agricultural and residential area	Pb		Many plant species (please see paper)		Phytoextraction and phytostabilization	*T. minuta* and *B. pilosa* were able to accumulate more Pb than other studied plants and thus, suitable phytoremediation of Pb polluted soils	Salazar and Pignata (2014)
Mae Sot contaminated fields, Mae Sot District, Tak Province, Thailand	Agricultural field	Cd		*Gynura pseudochina*, *Chromolaena odorata*, *Conyza sumatrensis*, *Crassocephalum crepidioides* and *Nicotiana tabacum*	2 months	Phytoextraction	All plant species, except *C. odorata*, demonstrated Cd-hyperaccumulating capacities and well treated most Cd-impacted soils at Mae Sot	Khaokaew and Landrot (2014)
"Gessenwiese", Ronneburg, Eastern Thuringia, Germany	Uranium mining site	U, Al, Ni and Zn	Calcareous topsoil, mycorrhiza and bacterial culture	*Helianthus annuus*, *Triticale* and *Brassica juncea*	14 months	Phytoextraction and phytostabilization	==Very low metal accumulation was reported in case of Triticale; *H. annuus* accumulated high Al whereas *B. juncea* accumulated high Zn and Ni in shoots; biogas and bioethanol production from biomass==	==Willscher et al. (2013)==
Carpiano, Milano and Torviscosa, Udine, Italy	Agriculture land	Cd, Cr, As, Zn, Pb, Co, Mn, Ni and Cu	Cement, lime, lime + iron sulphate, EDTA, EDDS, NTA, manure and biochar	Many plant species (please see paper)	10 years	Phytoextraction and phytostabilization	Chelators enhanced metals accumulation in plants from soil with increased biomass	Vamerali et al. (2012)
Torviscosa, Udine, NE Italy	Industrial waste contaminated area	As, Cu, Co, Pb, Zn and pyrite wastes		*P. alba* L., *P. nigra* L., *P. tremula* L., and *Salix alba* L.	2 years	Phytoextraction	Insignificant accumulation of As from waste, uptake of metals and As to above ground tissues is marginal and there is little risk of food web contamination *via* herbivores	Vamerali et al. (2009)
Lechang site, Lechang City, Guangdong Province, China	Mining site	Pb, Cd, Zn, and Cu	EDTA, NH_4NO_3 and $(NH_4)_2SO_4$	*Sorghum biocolor* L.	4 months	Phytoextraction	EDTA enhanced accumulation of Pb whereas NH_4NO_3 and $(NH_4)_2SO_4$ enhanced accumulation of both Zn and Cd in roots of sorghum plants	Ping et al. (2009)
Torviscosa, NE Italy	Industrial waste contaminated area	Cd, As, Zn, Pb, Co, and Cu		*Sorghum bicolor* and *Helianthus annuus*	40 days	Phytoextraction	Zn removal by *S. bicolor* and *H. annuus* reached about 2000 g ha^{-1} and 1000 g ha^{-1}, respectively	Marchiol et al. (2007)
Torviscosa, NE Italy	Industrial waste contaminated area	As, Cd, Cu, Pb and Zn		*Glycine max*, *Sorghum bicolor*, *Zea mays* and *Helianthus annuus*	50 days	Phytoextraction	*S. bicolor* appeared more efficient than *H. annuus* in terms of metals uptake	Fellet et al. (2007)

Fig. 1 Trophic transfer of toxic HMs from soil to plants to humans and organism's food to humans and their toxicity
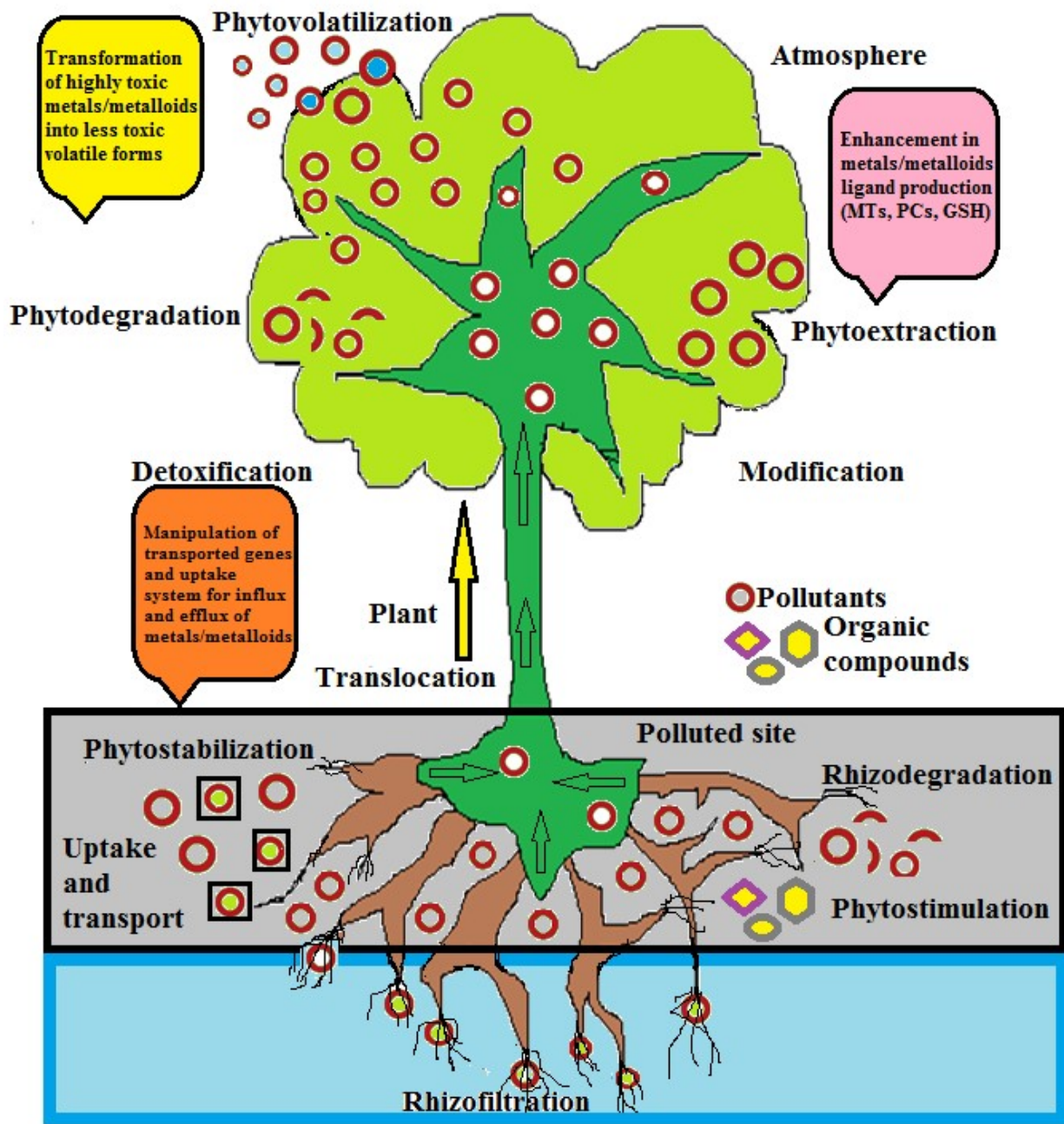
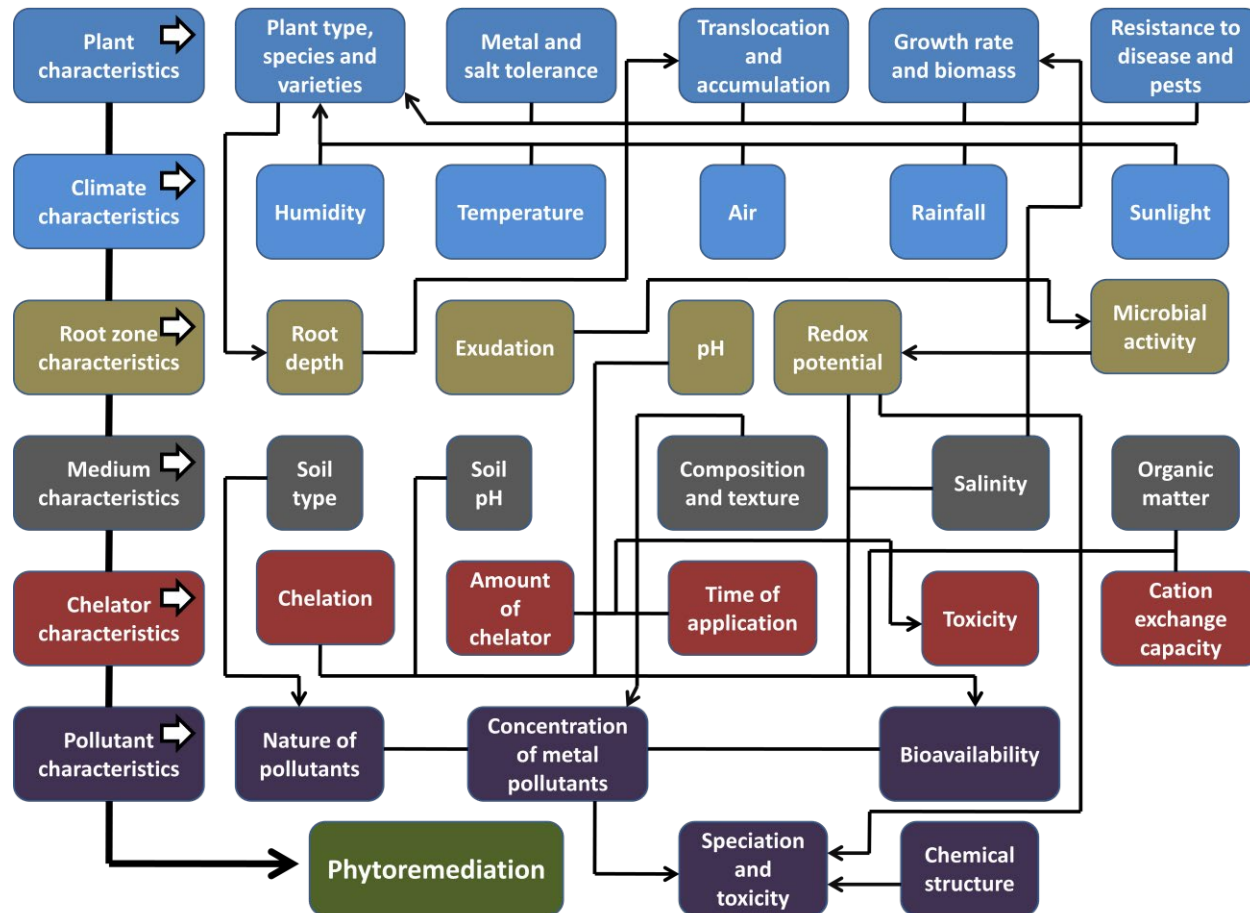**Fig. 2** A pictorial representation of different phytoremediation techniques

Fig. 3 Relationships among the factors affecting phytoremediation efficiency